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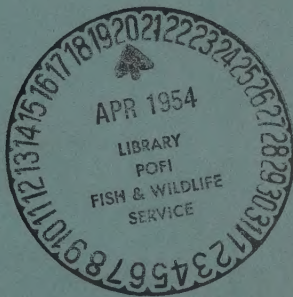
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THE SURFACE CURRENT FIELD IN THE WESTERN PART OF THE NORTH ATLANTIC¹

by

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ABSTRACT

By means of the equation of continuity, in the area chosen for the study, the divergence at each quadruple of square degree of latitude and longitude has been computed from the average surface currents, separately for all the four seasons and also for the whole year. On the righthand side of the axis of the Gulf Stream there seems to be a rather continuous area of divergence. This is shown to be consistent with the vorticity equation.

The area, the surface currents of which are studied in this paper, extends from longitude 48° W. to the Bahamas and the Atlantic coast of the United States and between the latitudes 19° N. and 40° N. The resulting surface currents for each month of the year in the chosen area were taken from the U. S. Hydrographic Office Charts No. 571, 1-12 for every square degree. For every square degree and for each month, the speed and direction of the surface currents were read from these charts. Every current vector was divided into two components.

The year was divided into four convenient seasons. When studying the annual cycle of surface currents in different regions of the area under consideration, it was found that the best way to select the different seasons was the normal, i.e., Winter: December-February; Spring: March-May; Summer: June-August; and Fall: September-November. (Fuglister, 1951, or Hela, 1952.) This division yields the minimum of fluctuation within any one season. After this decision the above components were taken into consideration anew. The sum and average for both of the components were computed for each square degree separately for each season of the year and similarly for

¹Contribution No. 119 from the Marine Laboratory, University of Miami.

the whole year. (Before accomplishing this step the order of magnitude of the resulting current for each month was compared with the other months and in the few cases with very distinct discrepancies the necessary corrections were made. The corrections were later checked and in the main approved by the U. S. Navy Hydrographic Office.) From these components, the resulting surface current has been computed for each square degree for every season (Figs. 3-7) and for the whole year (Fig. 8). The speed of the current is given in nautical miles per 24 hours. Those speeds found to be 0.5 nautical miles per 24 hours are indicated in the Figures by small circles only.

In order to evaluate the distribution of divergence in the surface current field, the equation of continuity has been used. The corresponding possibility has been used by Smith *et al* (1951) who indicated, by means of the stream function, (Pritchard, 1948) the distribution of vertical water movement from surface current vectors in the area of the Gulf of Mexico. The possibility of using the equation of continuity for this purpose is based on the following reasoning. The net outflow from a cube of unit volume is the sum of the three terms on the right of the following equation, and this outflow per unit time must equal the decrease of the density per unit time, $-\frac{\partial \rho}{\partial t}$.

$$-\frac{\partial \rho}{\partial t} = \frac{\partial(\rho v_x)}{\partial x} + \frac{\partial(\rho v_y)}{\partial y} + \frac{\partial(\rho v_z)}{\partial z}$$

This equation must therefore always be fulfilled in order to maintain the continuity of the system. On an average, in our case, the density of sea water, at least compared with the effect of the variations in the velocity, remains constant. Thus

$$-\frac{\partial \rho}{\partial t} \sim 0$$

and hence, with sufficient accuracy,

$$\frac{\partial v_x}{\partial x} + \frac{\partial v_y}{\partial y} = -\frac{\partial v_z}{\partial z}$$

Thus the horizontal divergence must be equal to the vertical gradient of the rate of upwelling (or downwelling). The effects of evaporation and precipitation can be taken as negligible.

The quantities

$$\frac{\partial v_x}{\partial x} + \frac{\partial v_y}{\partial y}$$

can be computed for every area *ABCD* (Fig. 1), if instead of *x* and *y* the directions SE and NE are introduced and if all the surface current

vectors are divided into their SE and NE components:

$$\frac{\partial v_{SE}}{\partial(SE)} + \frac{\partial v_{NE}}{\partial(NE)} = v_{SE,2} - v_{SE,1} + v_{NE,2} - v_{NE,1}$$

Positive values of these quantities correspond to the divergence (upwelling) and negative values to the convergence (downwelling).

The divergence has been computed for every intersection of the latitudes and longitudes in the area under consideration. Originally these computations were done for each month separately; however, the variations from point to point were great enough to suggest that the same ought to be repeated on seasonal basis. The results, without any smoothing procedure, are given in Figures 8-11 for the different

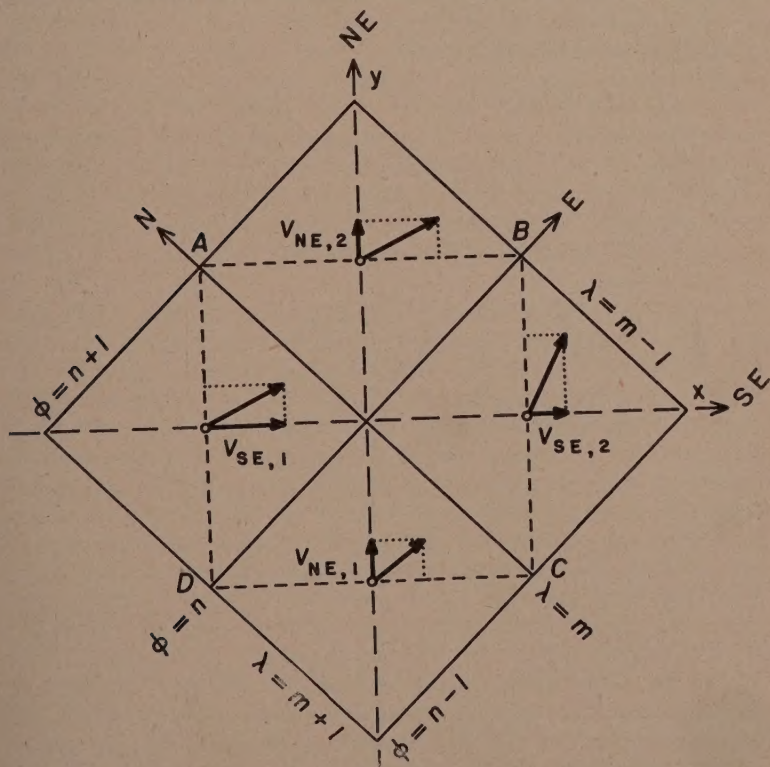


FIGURE 1. The computation of divergence on the basis of surface current vectors.

seasons of the year and in Fig. 12 for the whole year. (It is to be noted that in these drawings the negative values correspond to the divergence and the positive ones to the convergence.)

Certain characteristic details can be seen from the above Figures; however, several obviously impossible features are there, too. Especially in the area where the number of the current observations is rather limited, that is, outside the regular ships' routes, one exceptional current record can distort the distribution of the surface currents. In the distribution of the divergence, the same exceptional record may bring much greater local variations in the divergence than would correspond to the actual average distribution. In order to avoid this kind of distortion in the distribution of the divergence, the whole divergence field was smoothed by means of the formula

$$b'_2 = \frac{1}{16} [4b_2 + 2(a_2 + b_1 + b_3 + c_2) + (a_1 + a_3 + c_1 + c_3)]$$

where the symbols $a, b, c, \dots$ correspond to different latitudes and the symbols 1, 2, 3, $\dots$ to different longitudes. Thus b'_2 is the smoothed value of divergence in the place of b_2 , that is, at the latitude b and at the longitude 2. The results of this smoothing procedure can be seen in Figures 13-20 for the different seasons and in Figures 21-22 for the whole year.

In order to avoid obviously misleading current vectors in the final drawings, the current distributions of different seasons were also smoothed. This was done by computing both the mean speed and the mean direction of the current in each quadruple of four square degrees and letting the result correspond to the current in the area of the common point of the square degrees in question. It was seen that the deviations in the direction of the current were practically nil in most cases. Therefore the arithmetic means of the speeds were considered sufficiently accurate for this purpose. There were only two types of exceptions to this regularity. (1) At the northernmost edge of the Gulf Stream there was, of course, a distinct discontinuity in the direction of the surface current. The mean currents were not computed at all for the quadruples with currents flowing in different directions. (2) In the middle portion of the Sargasso Sea with weak currents, the directions seem to vary quite arbitrarily. It was found that if at least one of the four current vectors in question was less than 1.5 nautical miles per 24 hours, the directions did not, in general, coincide with each other. Therefore, in these cases the current vectors have been omitted in the final drawings.

In order to explain Figures 13-22 the following remarks are necessary. It was not possible to indicate the positive and negative divergences in the same charts clearly enough. Therefore the representations of divergence and convergence have been drawn separately, though the corresponding charts are, in other respects, identical in pairs. The areas in the middle portion of the Sargasso Sea, as already mentioned, where the currents are not indicated by arrows in the Figures, correspond to the areas where the resulting currents are about 1.5 nautical miles per 24 hours or less. The broken lines give the border of the speed 5 nautical miles per 24 hours. This border line can be seen along the northern edge of the North Equatorial Current, along the northeastern edge of the Antilles Current and along both the edges of the Gulf Stream. It is a common experience that both the direction and the speed of a monthly resulting surface current vector are uncertain if the displacement is small, because an astronomical fix is usually not accurate within 1 or 2 nautical miles, and the accuracy of a position by dead reckoning is, as a rule, less. Therefore, in most cases, little weight can be given to displacements of 5 nautical miles or less per 24 hours. In spite of this experience, our Figures 3-12 show that the seasonal and annual resulting currents computed from the ships' logs seem to be representative of the real resultant currents even in the case of weaker currents, *e.g.* 1.5-3 miles per 24 hours. As to the steadiness (or variability) of the vectors, it would have been of great interest to add the representation of steadiness to that of velocity and direction of the current charts. Unfortunately, we did not have any way of computing the steadiness of the surface currents for every square degree.

On the basis of surface current charts, average surface currents in several areas have, in the past, been represented by means of streamlines and equiscalar curves of speed. The principal advantage of this cartographic representation is that it permits a rapid survey of the major features and that it brings out the singularities of the stream lines. (W. Bjerknes, 1911). In our Figures the stream lines are not drawn due to the simple fact that they would have made the Figures hard to study. Nevertheless, the arrows indicating the current vectors might make it easy for anyone interested to determine the paths of the stream lines. In this connection it is essential to emphasize that representations of the actual surface current field must be based upon a very large number of measurements. These measurements must be simultaneous or, if not, it is necessary to assume that

the vector field is stationary. In this paper average surface currents are considered. The current vector field is, strictly taken, certainly not stationary; however, the result even in this case seems to show that the average surface currents, especially when computed on a seasonal basis and by means of a great amount of data, have a clear significance. As an example, for the surface current studies on the area also considered in this paper, the study of Felber (1934) should be mentioned, among others, in which the average surface currents, separately for each month, are indicated by stream lines.

The axis of the Gulf Stream is indicated by a heavy line with arrow heads on it. The figures beside the heads give the mean speed of the current on the axis in nautical miles per 24 hours. In addition, the areas of positive and negative divergence are separated with heavy lines. The areas themselves are cross-hatched with lines corresponding to the latitudes and longitudes. A smoothed value of divergence has been computed, as already mentioned, for each crossing point of the lines in question. The magnitude of (positive or negative) divergence is indicated by Figures 1, 2, 3, . . . and by corresponding isolines. Originally these Figures refer to the equation of continuity; however, they can be interpreted in the following way. The mean superficial magnitude of an area similar to *ABCD* is in the region of this study about 2.1×10^{14} cm². The value of divergence in the above scale corresponds to a mean current, through one of the four sides of *ABCD*, having the speed of 1 nautical mile per 24 hours, or 2.1 cm s⁻¹. We can roughly assume that the layer in which the phenomena connected with the singularities occur has on an average a thickness of 70 meters. The sides of *ABCD* are on an average 145 km long. Therefore, water is subtracted from the area *ABCD* with a rate of 2.2×10^{11} cm³ s⁻¹. If we assume that the corresponding upwelling occurs in the whole area *ABCD*, the value of divergence corresponds to the mean rate of upwelling 1×10^3 cm s⁻¹, which seems to be a relatively reasonable value; e.g., McEwen (1934) computed the rate of upwelling in the region of San Diego to be about 20 m a month, which corresponds to 0.8×10^3 cm s⁻¹.

In Figures 13-22 several interesting features can be seen, some of which are already well known. At the northern edge of the North Equatorial Current the mean direction seems to be toward WNW during winter, and toward NW or NNW during summer. During the summer the 5 mile curve seems to be shifted 200 or 300 km north. Even these representations seem to show that the Antilles Current

is only a relatively weak current. The 5 mile curve runs relatively parallel to the eastern edge of the Bahamas Banks and mostly rather close to it. In addition to the regular Antilles Current close to the Bahamas, there seems to be another current branch, coming from the North Equatorial Current and bringing water into the Gulf Stream approximately along the 70° W. In the region of 30° N. and 79° W. there seems to be a divergence area throughout the year which probably could be explained, at least to a certain extent, simply by the influence of the bottom configuration. The Antilles Current seems to join the Florida Current in the area of 31° N. and 78° W. In this area a strong convergence is seen.

Perhaps the most interesting features are seen along the Gulf Stream. The mean location of the axis of the Stream remains very accurately in the same area from season to season. This seems to be the case at least up to the 64° W. in the east. The speeds vary along the axis in the manner already well known.

On the righthand side of the axis of the Gulf Stream there seems to be a rather continuous area of divergence. The maximum divergence occurs about 200 km to the right of the mean location of the axis. Furthermore, this divergence seems to be most pronounced in the following three regions: (1) 33° N., 75° W.; (2) 37° N., 70° W.; (3) 38° N., 65° W. These three divergence maxima are persistent throughout the whole year; in fact, during the spring, the continuous divergence area is broken down into three areas corresponding to these.

The existence of the divergence band on the right side of the current axis is consistent with the vorticity equation. The individual change of the relative vorticity, in a horizontal field, may be expressed (See, e.g., Bjerknes, 1951, page 582, or Raethjen, 1953, page 180) by

$$\frac{d\lambda}{dt} = -\frac{dl}{dt} - (l + \lambda) \operatorname{div}_H v + \left(\frac{\partial R_y}{\partial x} - \frac{\partial R_x}{\partial y} \right) + \frac{N}{F_H} + \eta$$

where λ is the relative vorticity

$l \equiv 2 \omega \sin \phi$ is the vorticity of the earth,

v is the velocity,

R is the total effect of frictional forces,

$\frac{N}{F_H}$ is the density of solenoids, and

η describes the influence of the vertical motion in changing the vorticity about the vertical. The terms $\frac{N}{F_H}$ and η can be neglected because of their smallness.

The conservation of the absolute vorticity is given by

$$\frac{d}{dt}(\lambda + l) = 0$$

and, therefore,

$$(l + \lambda) \operatorname{div}_H v \sim \left(\frac{\partial R_y}{\partial x} - \frac{\partial R_x}{\partial y} \right).$$

Finally we may take the y -axis in the direction of the current and, relatively accurately,

$$\zeta \operatorname{div}_H v \sim \frac{\partial R_y}{\partial x}$$

At the lefthand edge of the Gulf Stream the change of the frictional effect, when moving into the positive x -direction, is negative. Therefore also the divergence term has to be negative since the vorticity is positive. At the righthand edge of the Stream the corresponding change of the frictional effect is positive and also the divergence term is positive. Thus it seems probable that the appearance of divergence band on the right side and of the convergence on the left side of the Stream axis are essential features of the mechanism of the Gulf Stream in this area.

It is worthwhile trying to find some compatibility between the above and earlier results. Wüst and Defant (1936) have given the average distribution of the subsurface temperature, salinity, and density in the North Atlantic as an Atlas to Vol. VI of the German "Meteor" Expedition. At a depth of 200 meters, a distinct tongue of high salinity south of the Gulf Stream seems to coincide with the above divergence area. The same is the case in the corresponding charts of the recent work by Fuglister (1953). (Also, the location of his 200 meter depth temperature anomalies, $>2^\circ$, on the right side of the Gulf Stream follows the region of our divergence region rather closely.) Moore (1952) has given the distribution of extinction coefficients in the North Atlantic Ocean. In his chart No. 15 there is a very pronounced extinction coefficient minimum in the area corresponding to the above regions (2) and (3).

On the lefthand side of the axis of the Gulf Stream there is an area of convergence. This distribution of the divergence and convergence would correspond to the schematic picture given in Figure 2. In this area the axis of the Gulf Stream seems to follow the line between the convergence and divergence areas or seems to be perhaps more closely a little on the side of the convergence. Actually, there is probably a slight component in the current arrows through the axis from right to

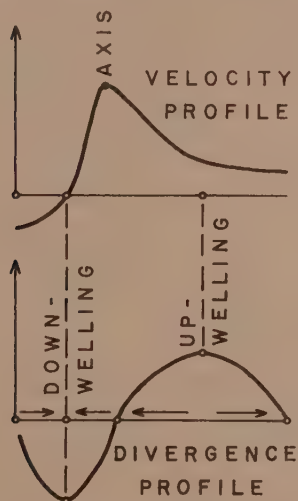


FIGURE 2. The schematic velocity profile and the corresponding distribution of divergence (and convergence) along the axis of the Gulf Stream.

left, as can be seen from Figures 13-22, or, if we assume that the directions of the currents on the righthand side of the axis and along it would be parallel to the axis, the divergence distribution has to be interpreted as follows: The existence of divergence shows that the speed of the waters running on the righthand side of the axis increases faster than the speed decreases at the axis. This simply means that water is added to the Gulf Stream from the area right of the axis.

In the area of the Sargasso Sea, convergence is seen, as expected, almost all over the area. However, certain limited divergence areas seem to appear in the area from which the one along the 70°W. is perhaps the most persistent (*cf.* Fig. 22). The reality of the other scattered divergence areas is probably questionable.

In addition, it is worthwhile mentioning that in the season to season changes of the surface current field some interesting details can be seen.

Acknowledgment. The author wishes to thank Dr. Erik Palmén for his valuable suggestions, and Mr. Frank Chew and Mr. Paul Ferris Smith for their critical remarks.

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Legend for the figures 3-12 on pages 251—260.

FIGURES 3-7. The resultant surface currents during different seasons and the whole year. The speeds are given in nautical miles per 24 hours.

FIGURES 8-12. The computed values of divergence of the average surface current field for winter. (In these figures the negative values correspond to the divergence and the positive ones to the convergence.)

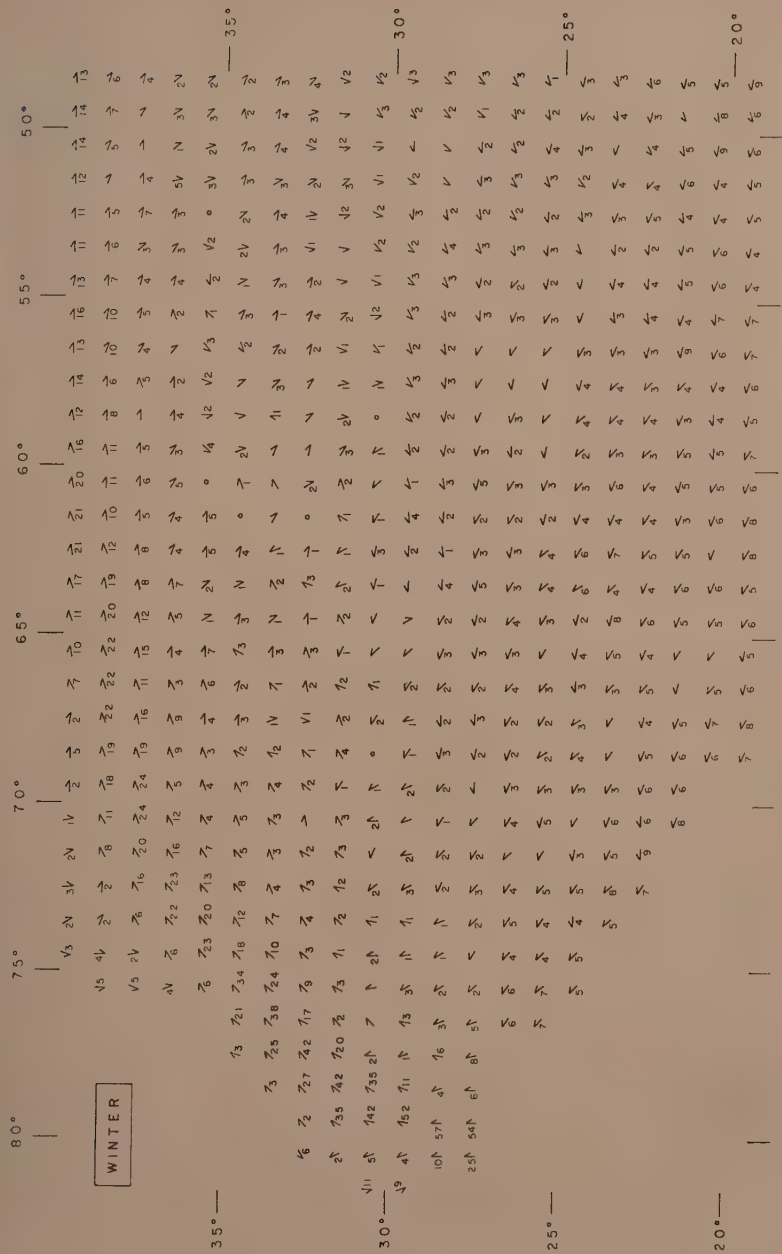


FIGURE 3.

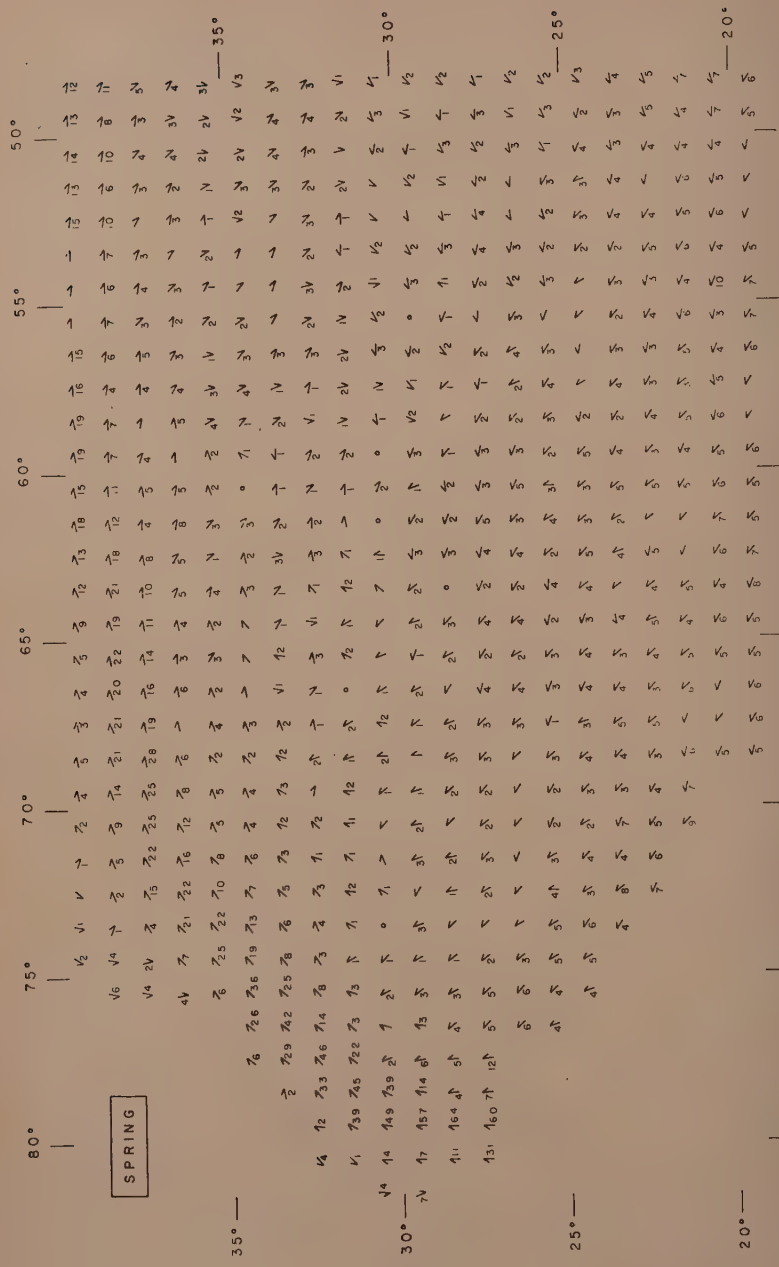


FIGURE 4.

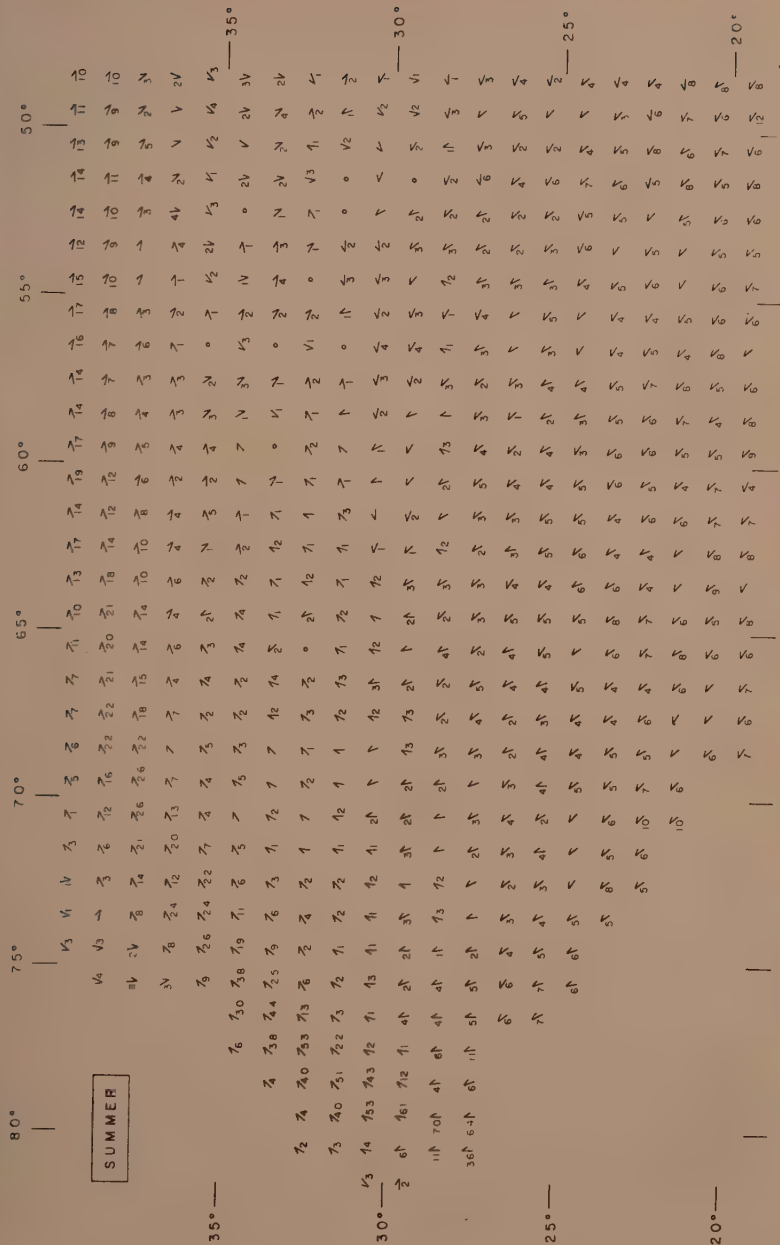


FIGURE 5.

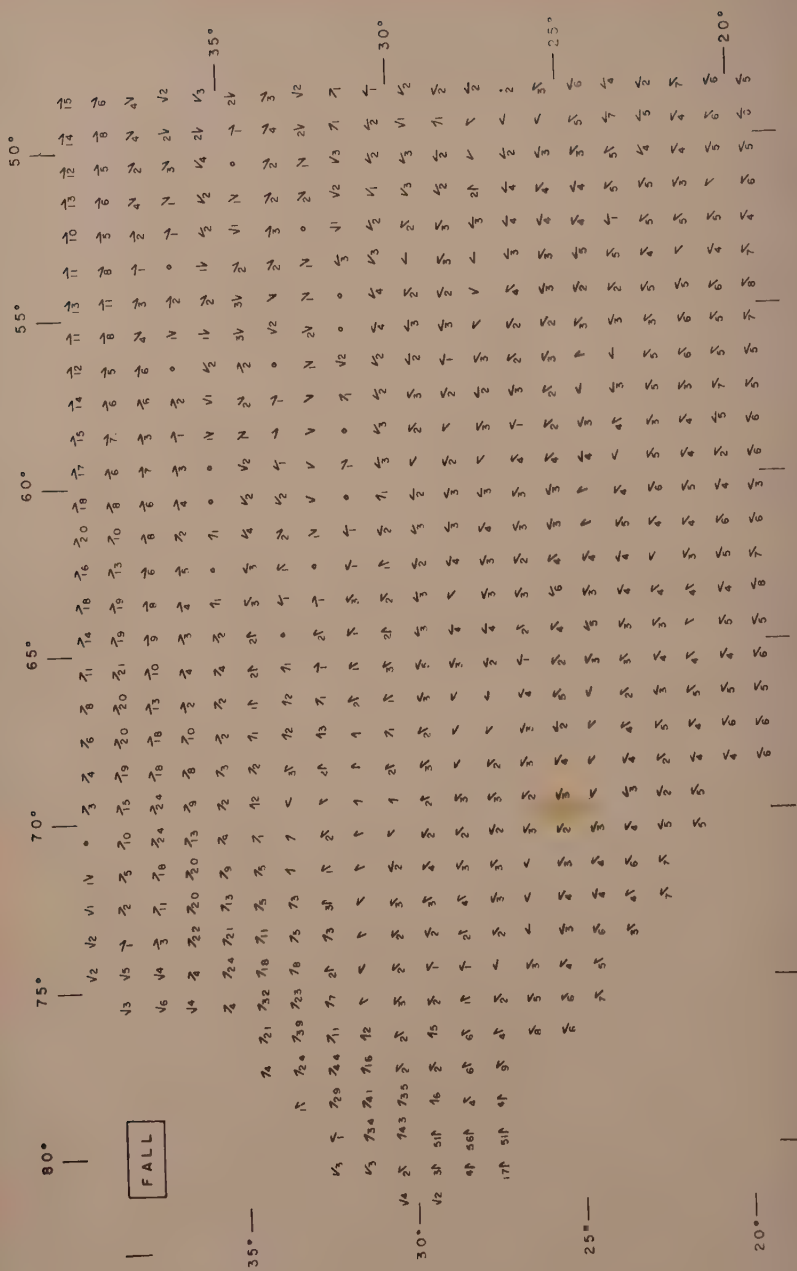


FIGURE 6.

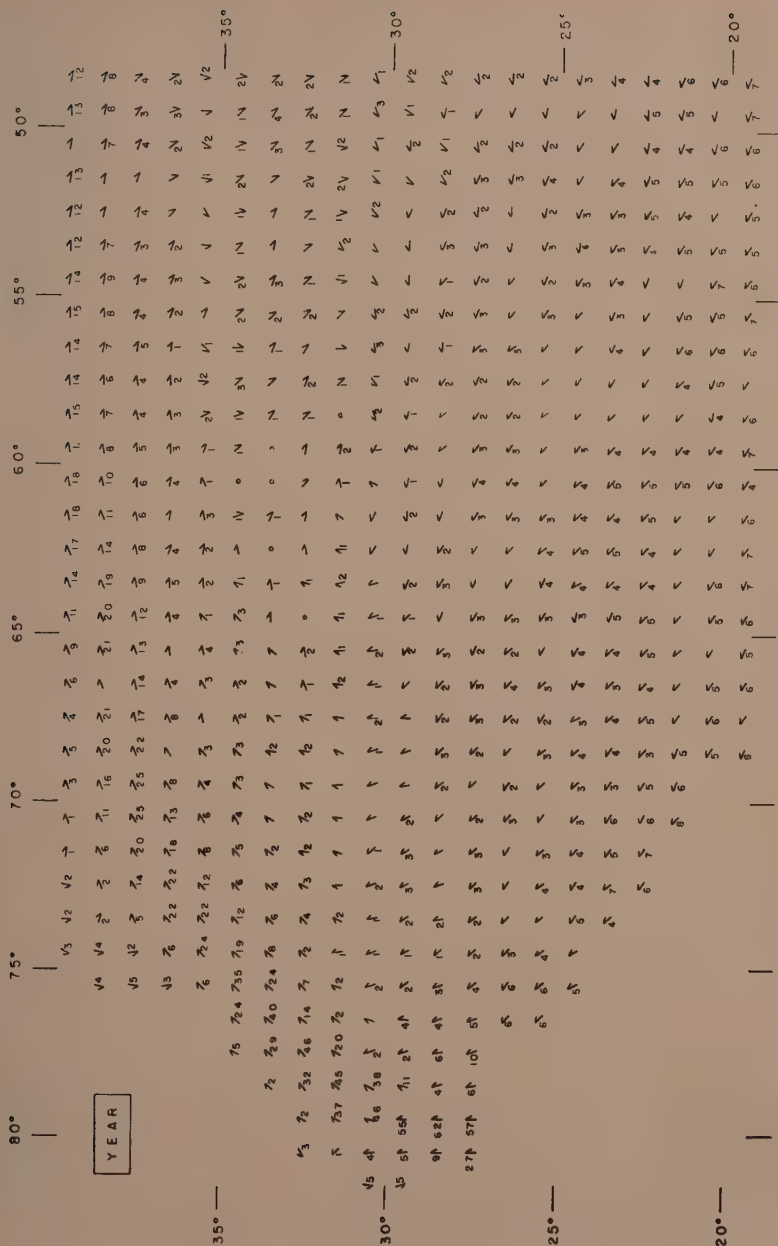


FIGURE 7.

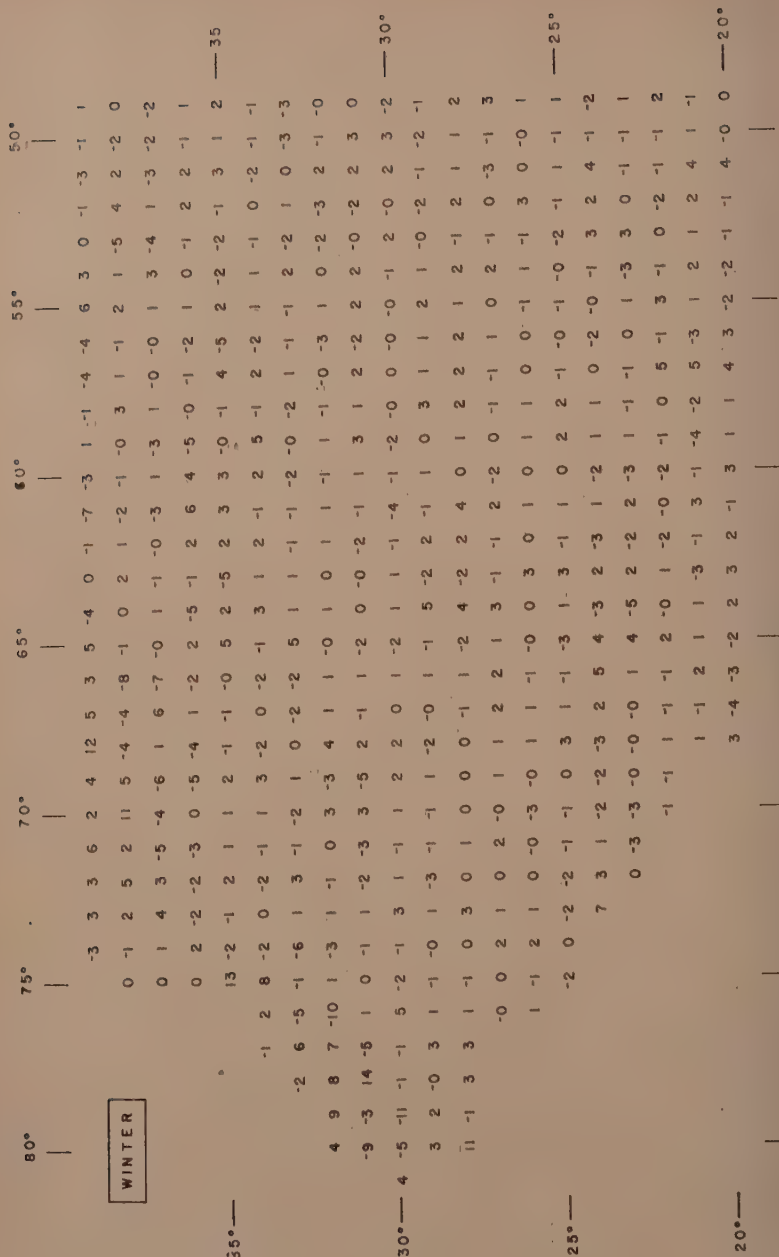
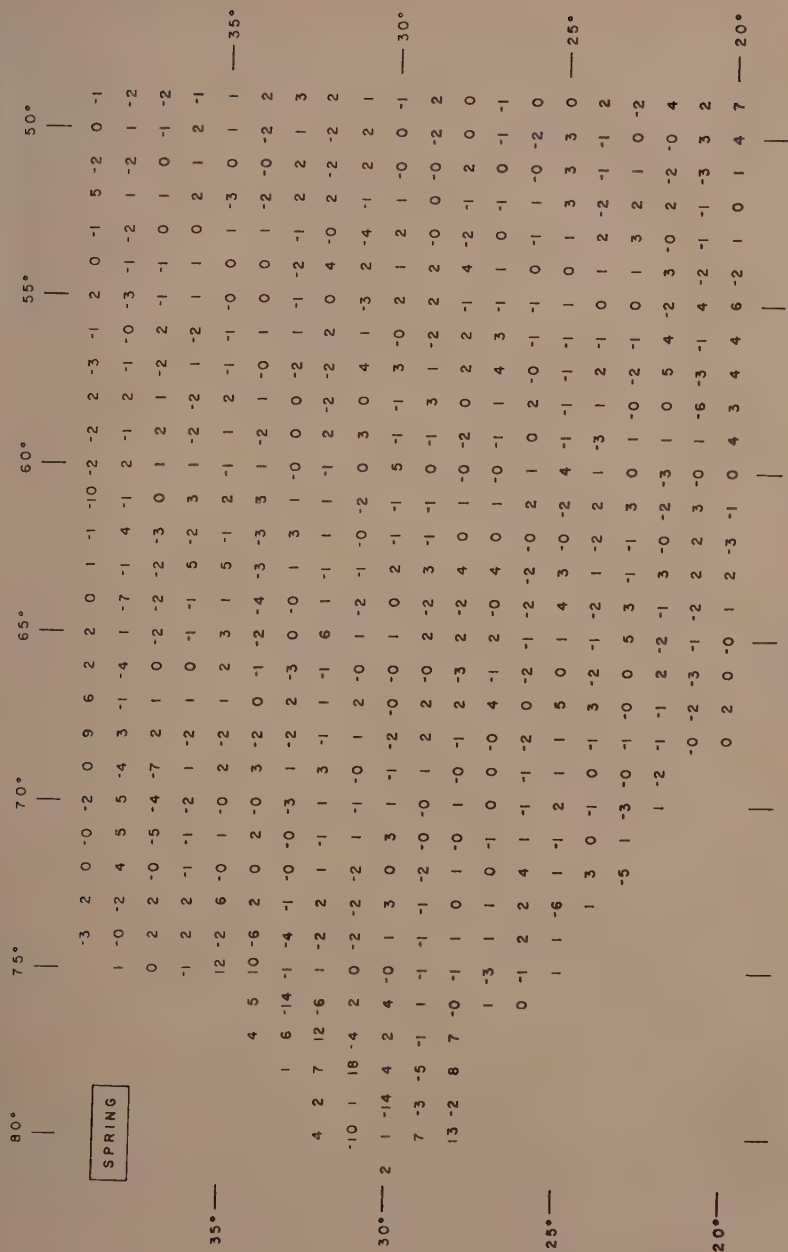


FIGURE 8.



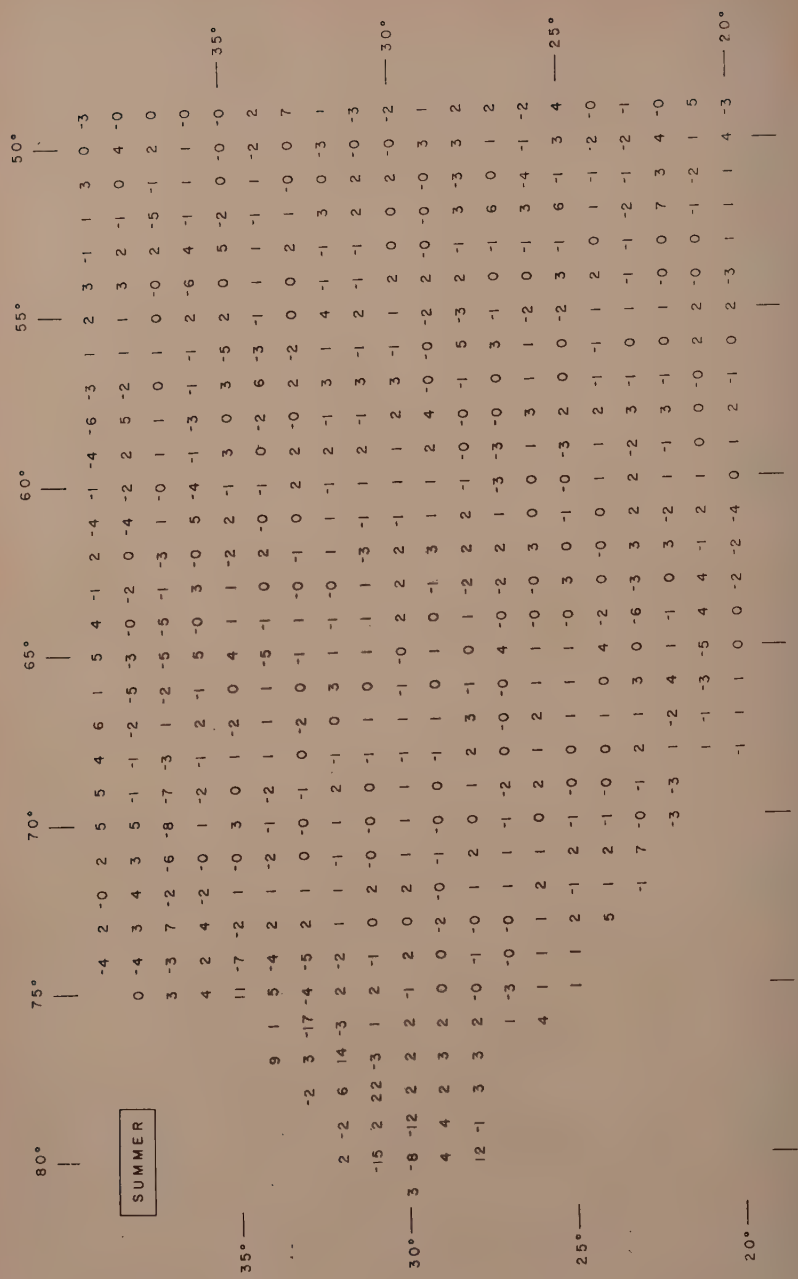
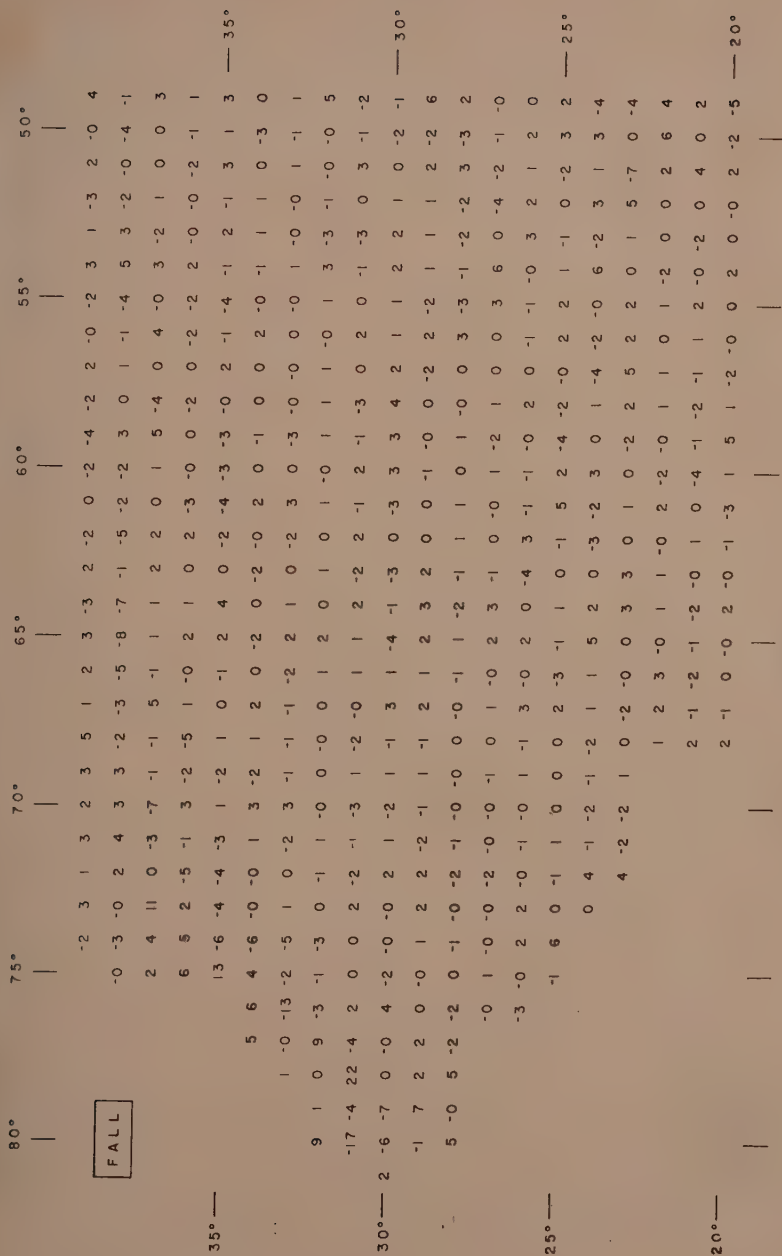
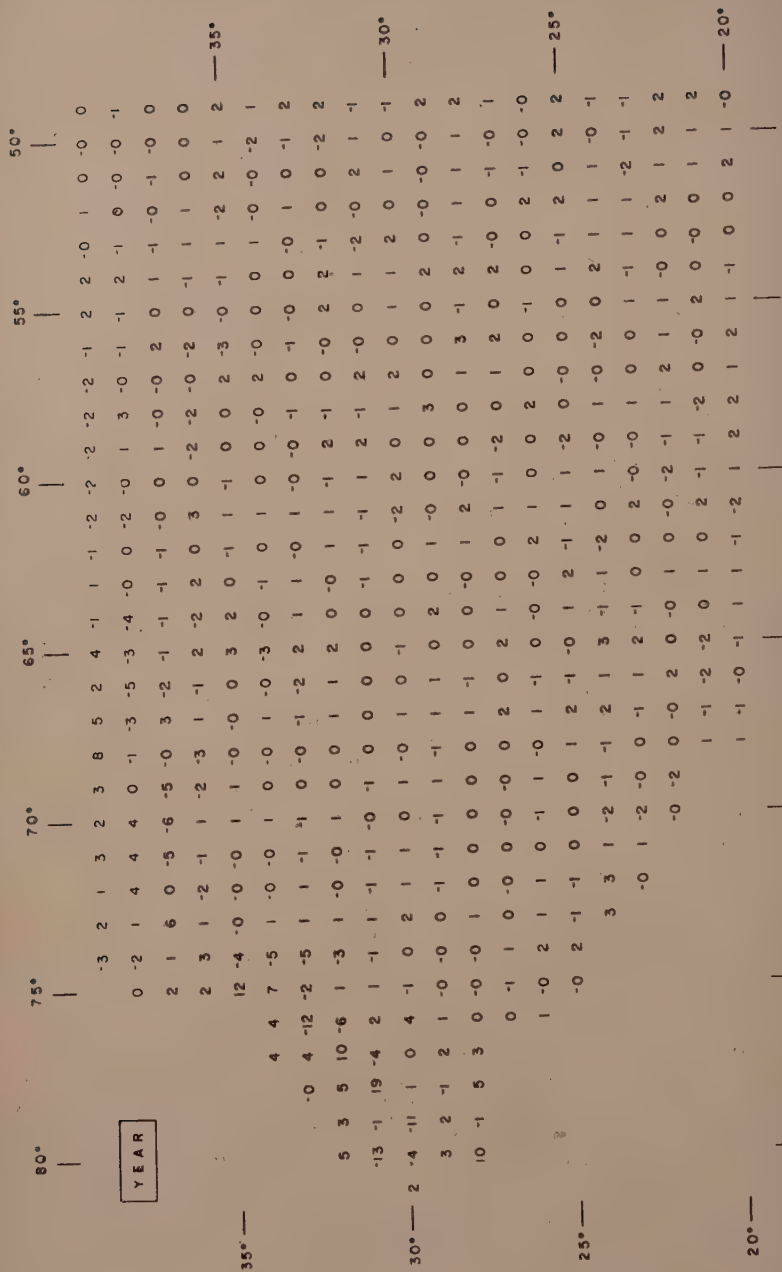


FIGURE 10.





Legend for the figures 13-22 on pages 262—271 L.

FIGURES 13, 15, 17, 19, and 21. The average surface current field in the western part of the North Atlantic during different seasons and the whole year.

The dotted lines correspond to the 100 fathom depth curve. The axis of the Gulf Stream is indicated by a heavy line with arrow heads on it giving the mean speed of the current on the axis in nautical miles per 24 hours. The broken lines indicate the 5 miles per day border of speed. The region with mean current speed of about 1.5 miles or less per day is not provided with the current arrows.

The areas with divergence are cross-hatched and the corresponding isolines indicate the mean rate of upwelling in $10^{-3} \text{ cm s}^{-1}$.

FIGURES 14, 16, 18, 20, and 22. The average surface current field in the western part of the North Atlantic during different seasons and the whole year.

The areas with divergence are cross-hatched and the corresponding isolines indicate the mean rate of downwelling in $10^{-3} \text{ cm s}^{-1}$.

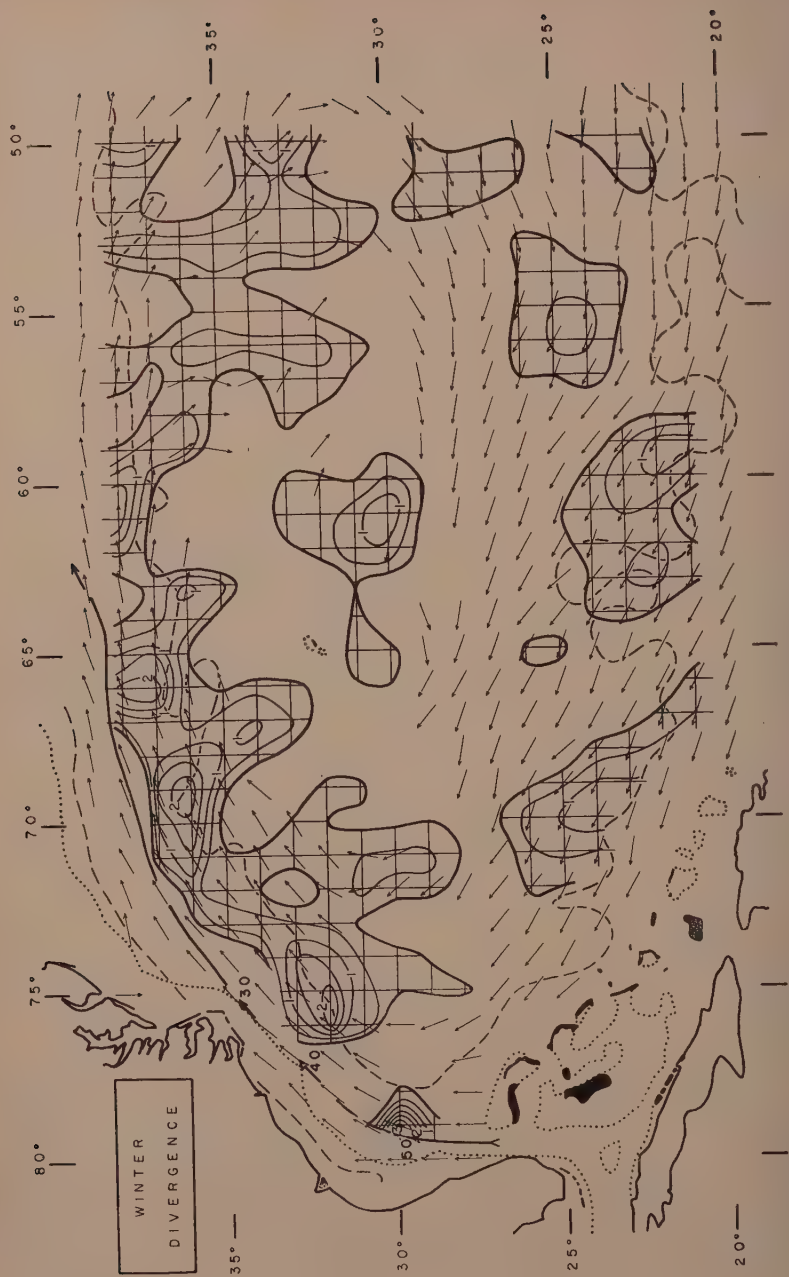


FIGURE 13.



FIGURE 14.



FIGURE 15.

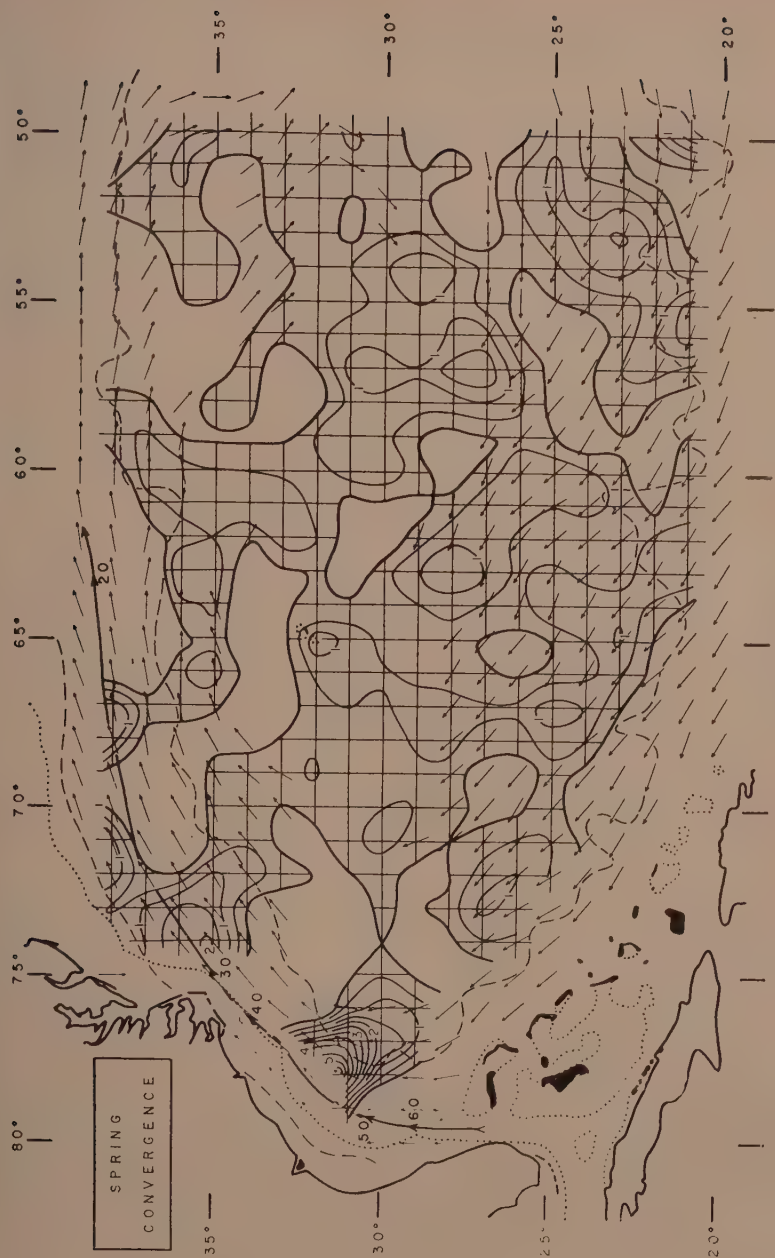


FIGURE 16.

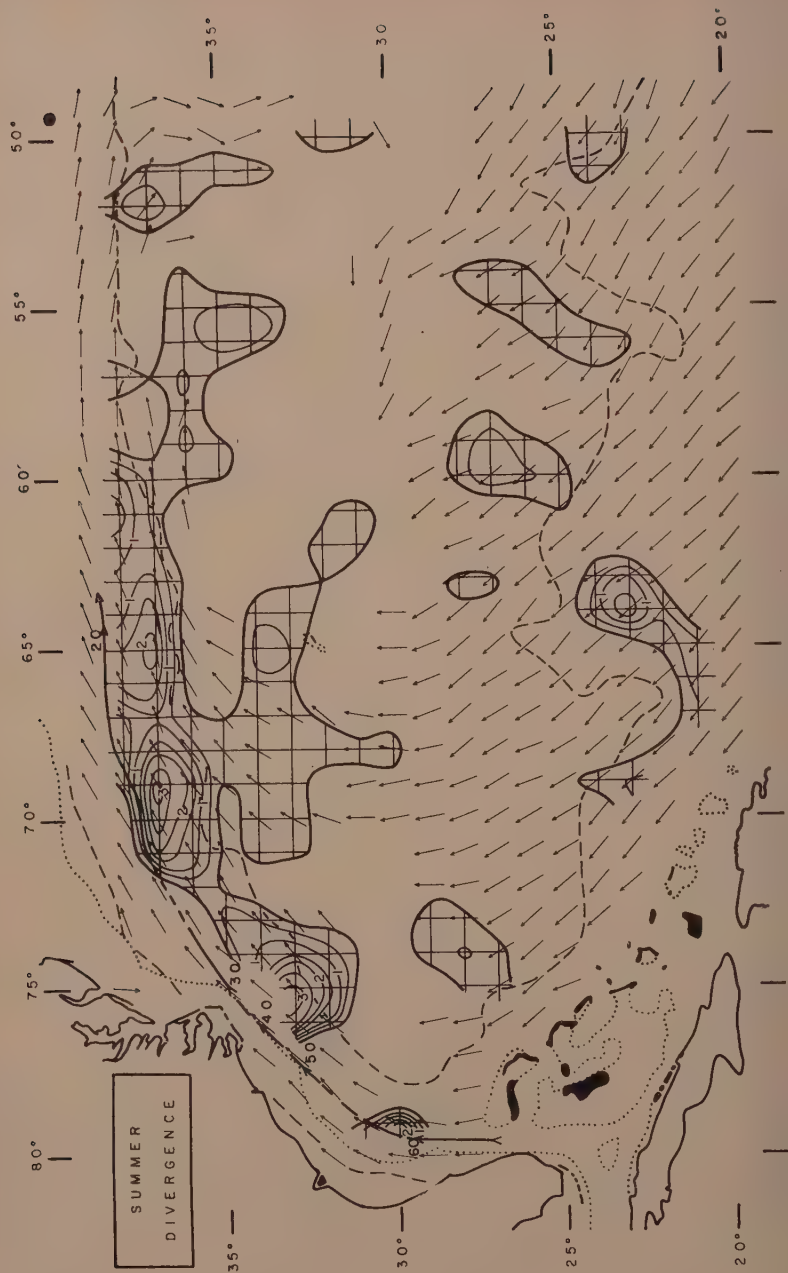


FIGURE 17.

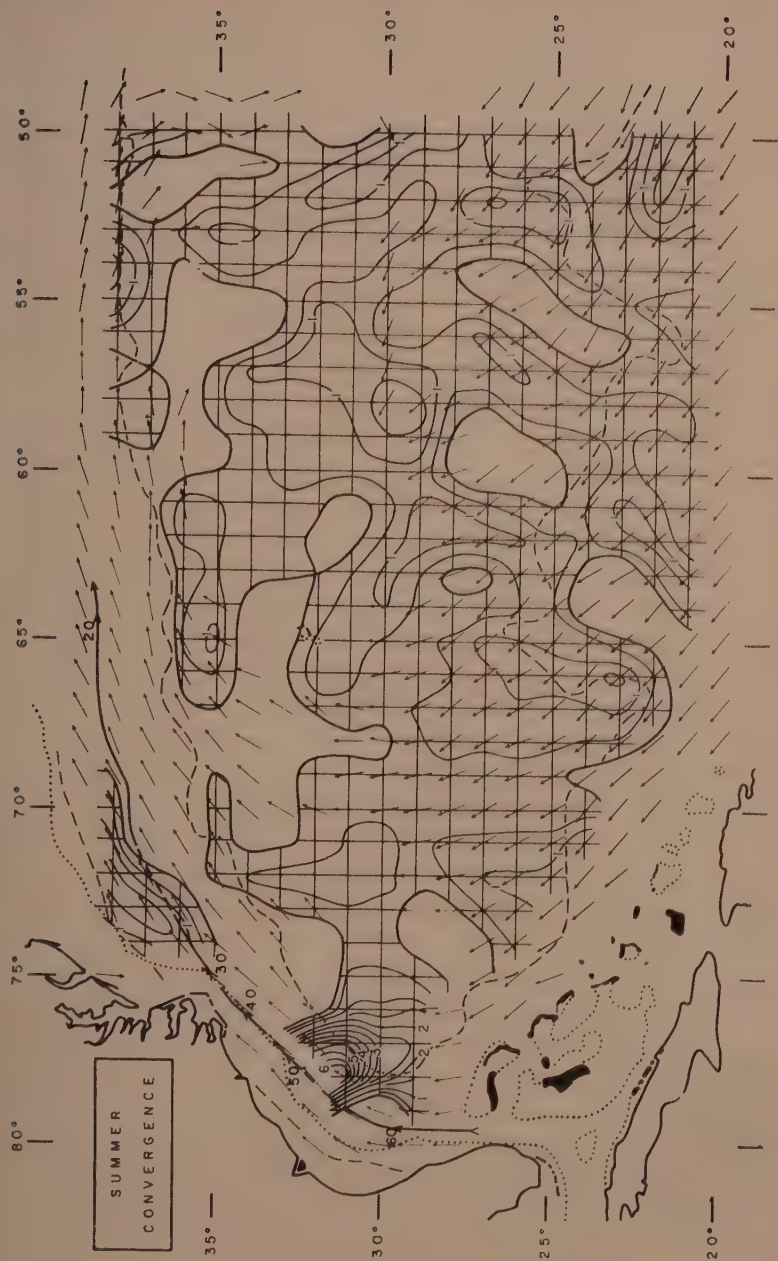


FIGURE 18.

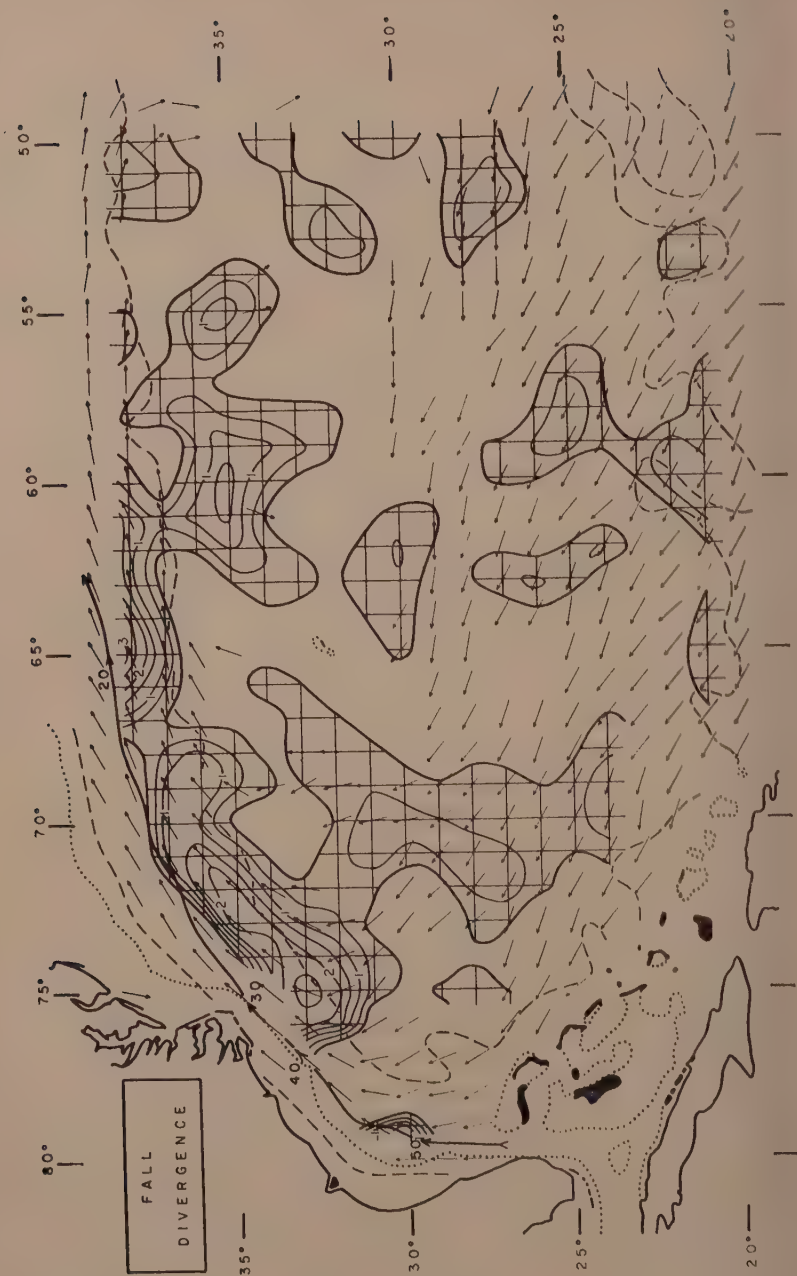


FIGURE 19.

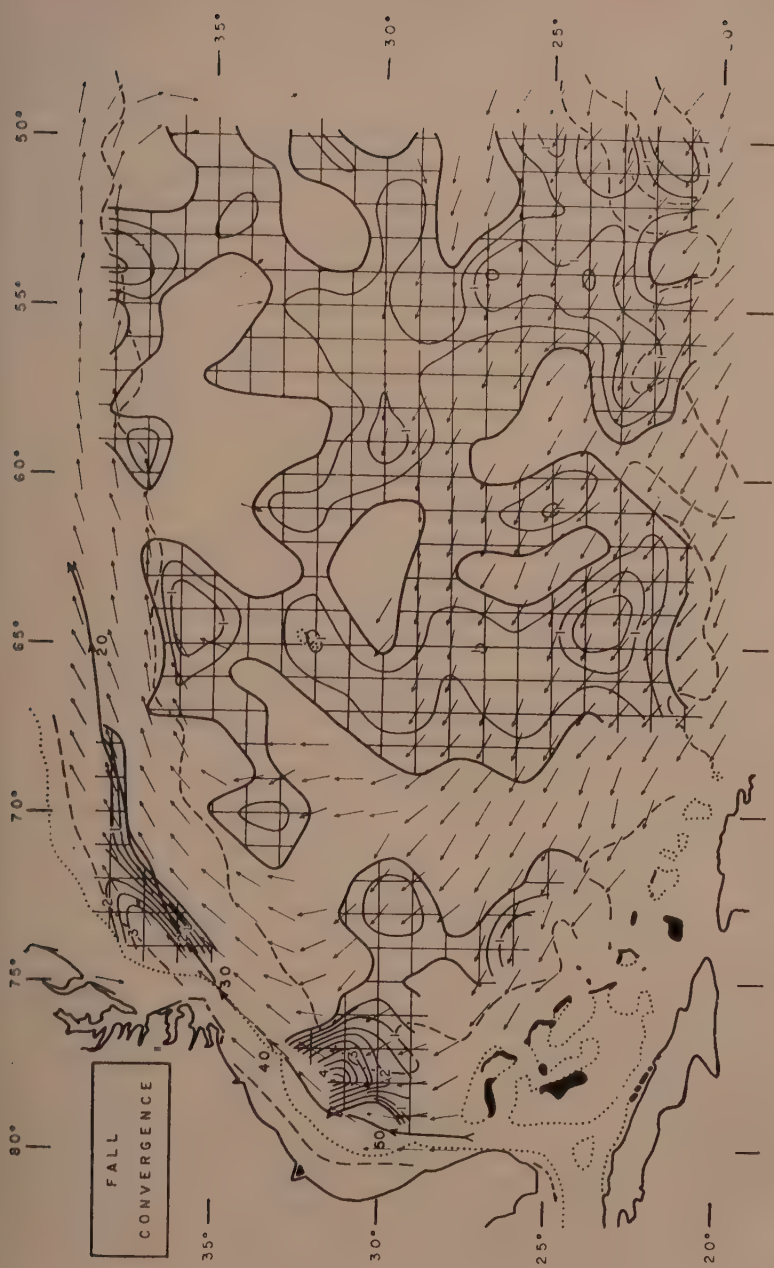


FIGURE 20.



FIGURE 21.

REFERENCES

BJERKNES, F.

1951. Extratropical cyclones. *Compendium of Meteorology*, Amer. Met. Soc., Boston, Mass. pp. 577-598.

BJERKENS, V., *et al.*

1911. Dynamic meteorology and hydrography. II. Kinematics. Publ. Carneg. Instn. No. 88, pp. 1-175.

FELBER, OTTO-HEINR.

1934. Oberflächenströmungen des Nordatlantischen Ozeans zwischen 15° and 50° n.B. *Archiv d. Deutschen Seewarte*. 53. 1. Hamburg.

FUGLISTER, FREDERICK C.

1951. Annual variations in current speeds in the Gulf Stream system. *J. Mar. Res.* 10 (1): 119-127.
1953. Average temperature and salinity at a depth of 200 meters in the North Atlantic. Woods Hole Oc. Inst., Tech. Rept. No. 53-58, pp. 1-12. (Unpublished manuscript).

HELA, ILMO

1952. The fluctuations of the Florida Current. *Bull. Mar. Sci. Gulf and Caribbean* 1 (4): 241-248.

MC EWEN, GEORGE F.

1934. Rate of upwelling in the region of San Diego computed from serial temperatures. *Proc. Pan. Pacif. Sci. Congr.* 3

MOORE, HILARY B.

1952. Physical factors affecting the distribution of Euphausiids in the North Atlantic. *Bull. Mar. Sci. Gulf and Caribbean*. 1 (4): 278-305.

PRITCHARD, DONALD W.

1948. Streamlines from a discrete vector field; with applications to ocean currents. *J. Mar. Res.* 7 (3): 296-303.

RAETHJEN, P.

1953. *Dynamik der Zyklonen*. Leipzig. pp. 1-384.

SMITH, F. G. WALTON, *et al.*

1951. Distribution of vertical water movement calculated from surface drift vectors. *Bull. Mar. Sci. Gulf and Caribbean*. 1 (3): 187-195.

WÜST, G. AND A. DEFANT

1936. Atlas zur: Schichtung und Zirkulation des Atlantischen Ozeans. *Wiss. Ergebn. Dtsch. Atlant. Exped. 'Meteor'* Bd. VI, 103 Pls. Berlin.

SOME RECENT OBSERVATIONS ON THE BIOLOGY OF *LITTORINA ANGULIFERA* LAM. OF BISCAYNE AND VIRGINIA KEYS, FLORIDA.¹

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ABSTRACT

Recent studies of *Littorina angulifera* show a bilunar periodicity in spawning through ten months of the year. Sexual maturity is reached in about two years, or, for most snails, at a height of 15 mm. The greatest percentage of spawning occurs in those snails from 15 mm to 26 mm in height, with both spawning and copulation apparently linked with the occurrence of an inch or more of rainfall. Sperm are transferred in spermatophores. Pelagic eggs, embryos, and veligers are shed in succession to the amount of 760,000 per female in one spawning period. In the laboratory all develop to the veliger stage only but may remain alive for three weeks. Rate of growth of young snails (2.9 mm) continues to accelerate until a size of 8 mm is attained. After that, the rate of growth decelerates abruptly until a size of 14 mm is reached, after which deceleration is gradual for the remainder of the life of the animal.

INTRODUCTION

Thirty years ago the spawning habits of the familiar periwinkles were believed to fit neatly into a pattern that correlated with the position each held in the littoral zone. Tattersall (1920) summarized the facts for the species in Great Britain in this manner: *Littorina littorea* which lives in the Laminaria and Fucus zones, exposed only at extreme low water of springtide, lays egg capsules unattached and singly. *L. obtusata* (= *littoralis* L.) exposed at very low water, lays eggs in masses attached to weeds. *L. rudis* (= *saxatilis* L.) and *L. neritoides* live out of the water and are viviparous. Sewell (1924) in discussing the spawning behavior of *Littorina scabra*, an Indian species, showed that it occupies a position similar to the last two named, and like them spawns viviparously. In respect to the zonation of *L. littorea*, Moore (1935) found the lower limit to be mean low water of springtide but that the upper limit depends on the nature of local conditions. All required wetting at least from splash. It was later shown by Lebour (1938), however, that *L. neritoides* living above the high tide level does not reproduce viviparously but produces planktonic egg-capsules which hatch out as free-swimming veligers, and Lysaght (1941)

¹Contribution No. 115 from the Marine Laboratory, University of Miami.

showed that they were shed at the period of spring tides. Lebour (1945) also reported that *L. angulifera* of Bermuda living in the mangroves above the high tide level produces viviparously. In this paper the present observer will show that the *L. angulifera* of Virginia and Biscayne Keys, Florida, regularly produce eggs in the early part of each spawning period but veligers at the peak of this period. There is, then, in the forms which have been studied, a transition from oviparous to viviparous *Littorina*. The order might be: *L. littoralis* with eggs hatching as adults; *L. littorea* and *L. neritoides* with eggs hatching as veligers; *L. angulifera* producing first planktonic eggs, then veligers in each spawning; and, finally, *L. saxatilis* and *L. scabra* producing viviparously. The correlation with zonation is not as clear as was originally believed.

Wilson (1951) and Carson (1952) cite numerous cases of lunar periodicity in spawning, notably in the marine worms and mollusks. In speaking of *L. littorea*, Thorson (1946) mentions plankton samples that contained *L. littorea* larvae of two distinct spawnings and concludes that its spawning must be rhythmic. Any relation to lunar phase has not been considered. Lysaght (1941) observed that the egg capsules of *L. neritoides*, as noted above, were found in the plankton tows only at full moon and new moon, when the tide is high enough to cover or wash the zone in which the adults remain. Observations over a period of more than two years on the *L. angulifera* of Virginia and Biscayne Keys, Florida, indicate a bilunar rhythm in the spawning of this species, as will be shown below.

Besides the work on the breeding habits and spawning indicated above, the present observer has, in a limited way, studied the growth rate and population trends.

The author is indebted to the members of the Marine Laboratory of the University of Miami, especially Dr. Hilary B. Moore, for facilities and advice. Dr. Gairdner B. Moment, of Goucher College, has also generously provided facilities and material assistance that made possible the continuance of the work in Baltimore. For the many sample populations of *L. angulifera* which came to Baltimore the author is indebted to Mrs. Genevieve D. Miles, Craig Phillips, and Everett Jones. For all these kind services the author wishes to express sincere appreciation.

METHODS AND RESULTS

Sex differences in L. angulifera. To all appearances the shells of

male and female *L. angulifera* are identical. The apical angle of samples measured were found to be the same. A slight difference in the curvature of the lip of the male was noted. This makes it difficult to stand the empty shells upright. No such difficulty occurs with the shells of the females. Variation in color and shell thickness has not been found to be associated with sex. Growth studies (Lenderking, 1952) in which a marked and measured sample was collected and studied after eight weeks, indicate that the females grow faster than the males. This was also found to be true for *L. littorea* by Moore (1935) and for *L. scabra* by Sewell (1924).

Leidy (1845) has described the anatomy of *L. angulifera*. More recent study by Morrison (1950, unpublished) reveals some inaccuracies in the original work. This is notably true of the structure of the copulatory organ. This organ springs from a point below the right tentacle of the male. It consists of two cylindrical parts, the basal one of which is a third larger in diameter than the distal part. From the basal part a flat oval dark brown structure, light around the edge, extends ventrally about one half the distance of the diameter of the base. It is placed over the edge of the female shell during copulation. As pointed out by Morrison (1950, unpublished) the tubular part of the vas deferens terminates at the base of the copulatory organ, opening into a fold or groove along the dorsal side of this structure. This seems to be similar to the fold described for some prosobranchs of British waters by Fretter (1946). The penis in young snails (8 to 10 mm) is transparent to white, smaller in proportion, with a very inconspicuous clasper. With increasing size and sexual maturity the basal part and the projecting penis become quite large and a bright yellow color. The whole copulatory organ diminishes in size and color after mating.

The oviduct of the female lies along the rectum in the posterior part of the mantle cavity. It terminates in a projecting extremity just outside the rectum. Like the copulatory organ of the male, the oviduct is swollen and yellow at maturity.

When individuals of *L. angulifera* are placed in sea water they immediately move up and out of it. This has been observed by other workers in their ecological studies of *L. angulifera* at the Univ. of Miami Marine Laboratory (Bullis, Hutton, and Dow, 1950, unpublished). In fifteen samples of from thirty to seventy-five snails each, the present writer observed that the first half of each group to reach the top of the container was predominately male. This was particu-

larly true in May, October, and November when the first half of the snails leaving the water was 80% male. Since peaks in spawning occurred in these months the inference is that the females tend to remain near the sea water at the time when they release the spawn. Lebour (1948) noted a similar tendency of the snails to remain low on the mangroves during the breeding season in Bermuda but no mention is made of a difference in tendency of males and females.

The breeding season of L. angulifera of Biscayne and Virginia Keys. The present observer's work with these snails began in February of 1951. Eggs and veligers were first recovered in April. Work at the laboratory continued until early July of that year and included the summer months of 1952 and 1953. During the intervening months snail samples averaging fifty each (19 samples in 1951-1952; 12 samples in 1952-1953) of *L. angulifera* were received for examination and study. No snails were received in February, 1952, or January and February, 1953. The snails were found to spawn in every month of 1951 beginning with April, in every month of 1952 except January and March (no data for February), and in every month of 1953 except January and February for which there were no data. The two spawning periods of March, 1953, included only very small percentages of females. It is possible that the occurrence of spawning in March, 1953, when five samples in March, 1952, failed to show any spawning females, is due to the increased amount of rainfall in 1953. The length of the breeding season in this area is notably different from that in Bermuda which seems to be limited to July and August, as reported by Lebour (1945).

Frequency of spawning. The percentage of spawning females was noted for each of 16 samples examined from August, 1952, through July, 1953. The variation during this period may be seen in Figure 1 and Figure 2. The average of these sixteen samples, however, is 31.6%. Since this is close to one-third it may be assumed that mature females may spawn every fourth bilunar cycle.

Fifty-seven females that spawned at the full moon in June were marked with red cellulose paint and returned to the mangroves. Forty of these were observed on three successive days during the following new moon spawning period. None were found to have spawn. Of snails that were kept in the laboratory during the year none was observed to have spawn in any two successive periods. During the July full moon period, twenty-two of the original number were recovered. Of these 17 (77% of those recovered, 38.5% of

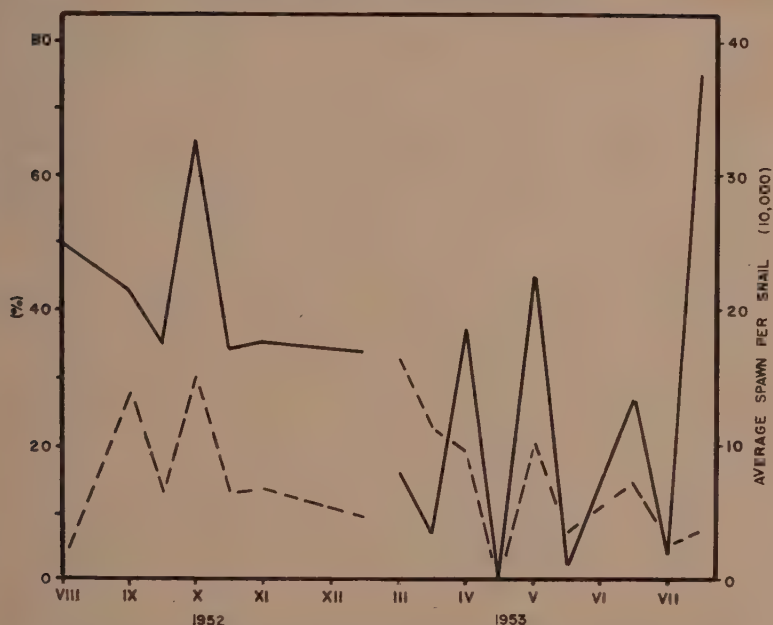


FIGURE 1. Seasonal variations in the percentage of females spawning (solid lines) and of the average number of eggs per individual (broken lines).

the original marked group) produced spawn. A random population taken at this same period also produced an unusually high percentage of spawning females, 75.7%. Only seven of the marked snails were recovered in the first August spawning period. None of these contained spawn in the mantle cavity. If conclusions can be based on these small numbers it would seem that the snails spawn in alternate lunar periods instead of the first and fourth as indicated by the laboratory count.

The unusually high percentage of spawning in the July full moon period and in the June full moon period were both preceded by above normal precipitation. The very low percentage of spawning in the July new moon period was preceded by a week of dry weather (0.02 inches of rain in eight days).

No precipitation record is kept for the Bear Cut area of Biscayne and Virginia Keys. The Weather Bureau, however, has a station on Miami Beach. Although this is about two miles north of the area in

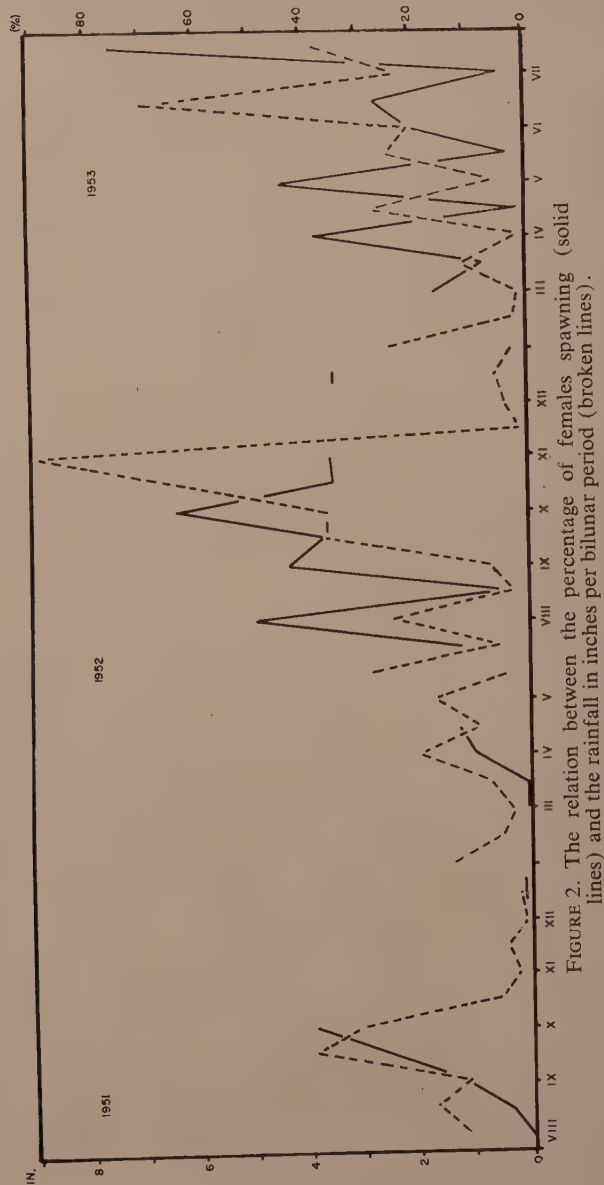


FIGURE 2. The relation between the percentage of females spawning (solid lines) and the rainfall in inches per bilunar period (broken lines).

question and local weather is variable, the precipitation here can be considered as a general index of the prevailing weather in nearby areas, particularly along the coast. The precipitation at this station has been totalled for each bilunar period and plotted together with the percent of spawning females in Figure 2. Though there is not a complete correlation, in general a period of low rainfall or diminishing rainfall is followed by or accompanied by a low or lower percent of spawning females, and about one inch of rainfall seems to be necessary to induce spawning.

Furthermore over a period of two years a peak in the spawning has been observed in both the spring and the fall months. An examination of the fifty year means for Miami show a gradual increase in rainfall from 1.8 inches in February to 7.2 inches in June. With a slight decrease in July (5.4 inches) it rises to 8.67 inches in October and drops to 1.8 again in December. Sewell (1924) observed that the peaks of the spawning period for *L. scabra* occur before and after the Southwest monsoon, linking the phenomenon with meteorological conditions.

It seems then that there is a possibility that *L. angulifera* females spawn eight times a year, assuming that spawning does occur in the months of January and February, or seven times a year if this is not true; that spawning does not occur in two successive spawning periods for any one female; that the frequency within the limits just described is determined by the degree of precipitation in the particular area under consideration.

Size of spawning females. In a population sample of over six hundred (3 mm to 26 mm in height), forty females were found spawning. The smallest of these measured 15 mm and the largest 26 mm. In fourteen smaller population samples examined during the year these were also found to be the limiting sizes. Seventy-five percent of the spawning individuals were found to fall in the 17 to 21 mm group. A random population sample usually has most individuals in this size group, too. In the course of these studies, however, at least two snails of 11 mm have been observed to produce spawn. One produced six eggs, the other fifty.

A more complete analysis of females in sizes 10 to 27 mm was attempted in the successive spawning periods of June and July, 1953. Of 409 females obtained within this range, only 5 were over 24 mm and a total of 18 for sizes 10 to 13 mm. These are not included in Figure 3 which shows percentage within each size group spawning.

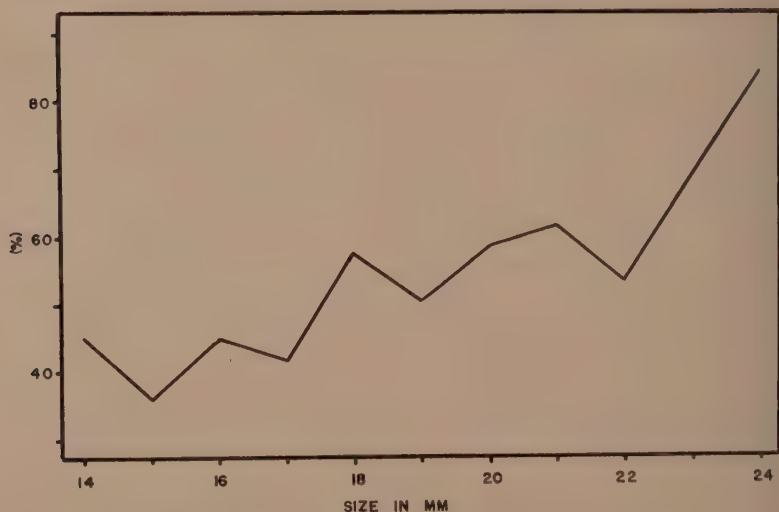


FIGURE 3. Percentage of spawning females in each size group.

None of size 10 mm was found with spawn. One or more of each of the other size groups did produce spawn.

Thorson observed (1946, page 422) that in several gastropods there is a direct relation between size of ovary and quantity of eggs produced. The small number of eggs produced by the two small snails mentioned above would seem to coincide with this. The relationship, however, is not evident in the larger sizes of *L. angulifera* as shown below.

Copulation. There is no size selection in mating *L. angulifera*. Very small males will be found copulating with very large females and vice versa. Most of the time the snails will be found scattered at random throughout the mangrove tree above water. Occasionally an isolated pair may be found in copulation at any time during the lunar cycle. They have been observed several days before the quarter phase of the moon and up to the day before full moon or the new moon. It is around the quarter phase of the moon, however, that pairing off becomes obvious. The number of pairs in copulation seems to be related to the amount of rainfall. On July 19, 1953, at the first quarter of the moon, a careful search of two trees where the population in June had been over a thousand, yielded only ten copulating pairs. On July 21, after a heavy thunder shower the previous night,

38 pairs were found on one of the two trees. In the previous year, following a rainfall of 1.55 inches the night before, over 90% of all the snails visible on the south side of Virginia Key were copulating. Similar observations were made in April, 1951, after a rainfall of 1.91 inches.

Pairs of snails have been observed on the mangrove trees with the male completely withdrawn but attached to the female shell with a dab of mucous. The female in these cases is firmly held to the tree with the muscular foot. In one group of ten pairs brought into the laboratory, copulating pairs were observed throughout two days, and in a second group for four days. In one case a pair remained in copulation for a half hour, in another for three hours. It seems that once a partner has been selected they remain together, the act of copulation occurring if moisture is optimum, being interrupted by increasing dryness only to be resumed again when conditions are more favorable. A slight touch, however, will remove the male from the female shell when attached by the dry mucous. It is likely then that each may have another partner if not several during the period of copulation.

Several males with well developed copulatory organs and several pairs of copulating snails were relaxed by placing them in the refrigerator over night (submerging them in distilled water by completely filling the jar also proved effective though use of drugs such as pilocarpin was not). By means of a capillary pipette a small amount of fluid was removed from the punctured vas deferens at the left side along the mantle. Some fluid was also removed from the dorsal groove of the copulatory organ. In both cases spindle shaped spermatophores in great abundance were recovered (see Figure 4). Shortly after these were mounted in sea water for examination under the microscope, some of the spermatids, the squarish structures arranged in rows along a central core, were found to develop into active sperm with very long lashing tails and minute heads. These same spermatophores were also found in the oviduct of the female after copulation. Myriads of sperm were found in the upper half of the enlarged oviduct where there were many small pale yellow spheres. The latter obviously were the eggs but lacked the hyaline case present in those recovered from the sea water.

Of three sets of copulating snails examined in July, 1952, (3rd, 14th, and 29th, respectively) eggs were produced on the fourth day after copulation. In July, 1953, eggs were recovered from both sets

on the same day though this was two days after copulation in one set and four days after copulation in the other. Since examination of fifteen to twenty-five females on successive days in the lunar cycle failed to show any spawn except at the new moon or full moon periods, spermatophores placed in the oviduct at earlier times in the cycle either were not effective in fertilization or were stored there until the eggs were ready. The presence of a sperm receptacle such

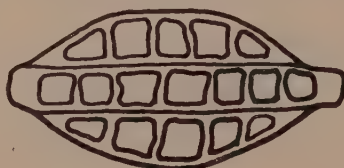


FIGURE 4. Spermatophore of *L. angulifera* ($10.6\mu \times 23.2\mu$).

as those described for other prosobranchs by Fretter (1946) is not obvious.

The number of snails copulating does not indicate the number that will produce spawn. For example, of ten copulating pairs observed July 3, eight females spawned; of eight copulating pairs observed July 14, one female spawned; of six copulating pairs observed July 29, three females spawned. Again, the relative amount of rainfall in these periods may be significantly linked with the successful production of spawn.

The Spawning cycle. At first spawning individuals were identified by putting the females in separate jars with a small amount of sea water. Sometimes the spawning female will go down the jar only far enough for the lip of her shell to be submerged. At other times she may crawl through the water over the bottom of the jar. The length of time she remains in the water at any one time varies from ten minutes up to an hour or more. Since the young were shed from any one female for a period of a week or more, it was concluded that the spawn was deposited and retained in the mantle cavity for that period. Dissection showed no pouch or sac but a circular mound of eggs covering the branchial ridges in the left dorsal part of the mantle cavity. Apparently the whole mass is held in place only by layers of mucous. The mass can be seen easily without dissection by simply tipping the shell of the live animal and looking into the mantle cavity. The mass may be grey, bright yellow or brown but at least in the early part of the period is most often bright yellow. Sewell (1924)

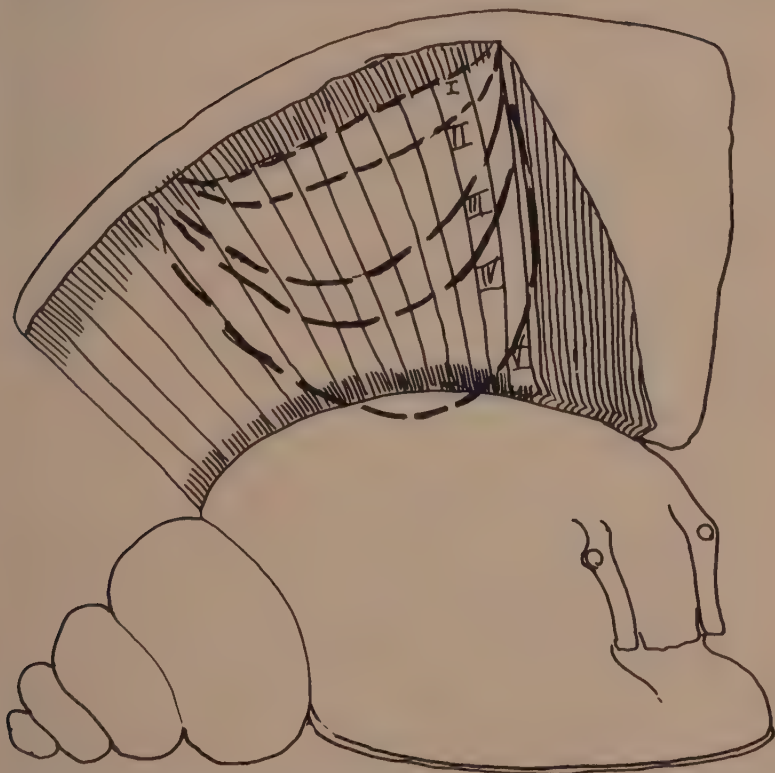


FIGURE 5. *L. angulifera* removed from the shell, and with the mantle reflected. The numerals indicate the successive boundaries of the regions occupied by the spawn during the deposition of the eggs.

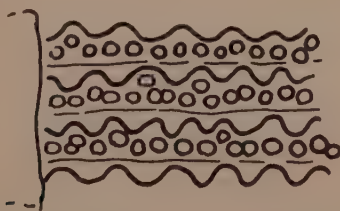


FIGURE 6. A portion of the mantle wall near the oviduct showing the eggs moving outward between the branchiae.

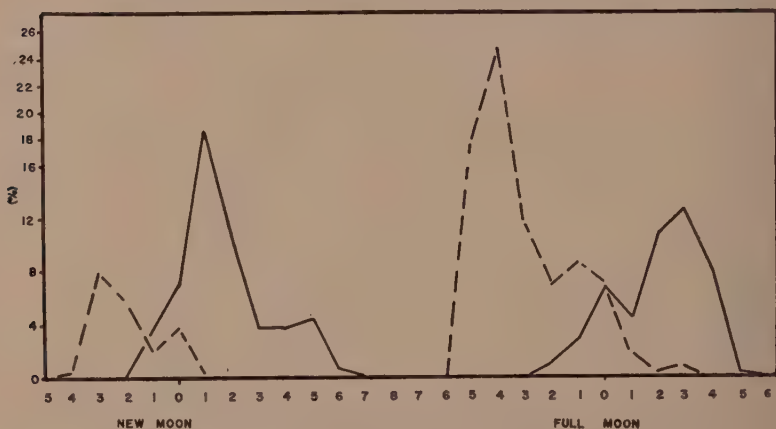


FIGURE 7. The occurrence of eggs or embryos, and veligers on successive days in the lunar month.



FIGURE 8. Developing egg of *L. angulifera*.

found that *L. scabra* carried the eggs in a similar position but he reports that the mass in this species is brown. The accompanying diagram, Figure 5, shows the mantle freed along the posterior edge and lifted from the right side to show the position of the egg mass. The eggs first appear in the mantle cavity along the edge of the rectum, then are moved outward between the branchia (see Figure 6). As additional eggs are deposited new layers slide over the first until the branchia are no longer visible.

This egg mass appears in the mantle cavity of the female between the quarter phase of the moon and the full moon or between the quarter and the new moon. The actual number of days before full moon or new moon varies from one to five. Any time following their appearance the eggs or embryos may slip down over the dorsal body wall, singly, in chain, or in rafts varying in number from six to over a thousand. The number of eggs or embryos diminishes as the full moon or new moon approaches and an increasing number of veligers

is released. The greatest number of veligers is released within a day or two of the full moon or new moon and the number drops sharply thereafter. All spawn is usually completely shed by the following quarter phase if not before. In making the counts in the laboratory, each snail was examined with a dissecting microscope to be sure all spawn had been released before the count was stopped. Since the total number of offspring per individual is so large, in preparing the accompanying graph, Figure 7, the eggs and embryos were totalled separately. The percentages stated are percentage of total of eggs and embryos per day and percentages of total veligers per day. Figure 7 shows very clearly the bilunar periodicity of the spawning. After spawning is complete there is often a large flocculent mass of white material discharged from the renal organ. Superficial examination might confuse this with the eggs. It is periodically discharged, however, by both males and females though rarely if ever by the latter during the spawning cycle.

The egg, pale yellow in color, is spherical and surrounded by a hyaline slightly ovoid case (see Figure 8). The eggs measured July 1, 1952, were 58.2μ inside diameter, 71.5μ with the hyaline covering. These eggs recovered at 11 A.M. had reached the four-celled stage by 2:45 P.M., the eight-cell stage by 4:00 P.M. By 11 P.M. the embryo was well advanced and had increased to 69.9μ inside, while the case measured 93.2μ in length and slightly under this in width and depth. Some of the eggs were kept in watch glasses, others were put in beakers with oscillating plates. Development proceeded in each case to the veliger stage. A check over a four day period showed the same stage of development in the dishes or beakers as those freshly obtained from the spawning female. Both those spawned as eggs or embryos and those spawned as veligers have been kept in the laboratory for as long as three weeks. Attempts to keep them longer have been unsuccessful probably due to inadequate food. Yeast cultures were used to encourage development of nanoplankton for food. Blue-green alga and dinoflagellates cultured in Allen and Nelson nutrient solution have also been tried. In no case has there been any appreciable change in size or structure noted in the veligers after the first day or so. The majority rest on the bottom of the container unless disturbed. The very small size and absence of stored food together with the above facts suggests that their pelagic life is of short duration. Thorson (1946, page 434) states for this type of larvae that feeding on plankton is of secondary importance while spread of larval stock

seems to be the main objective (advantage). He goes on to say that adaptability to unfavorable conditions is much greater than might be expected in such small larvae without any supply of yolk and no growth. He further states (page 454) that observations originating in nature show the pelagic life in some Littorinidae averaged two to three weeks. The present observer has noted that *L. angulifera* veligers may become fastened to the oscillating glass plates in small mounds of mucous near the water level. No further development was noted. It could be that the successful transition in the laboratory was not possible because of unsuitable substratum rather than inadequate food. Because of limited time the author was not able to check the presence in the plankton during the spawning period. The wide spread occurrence of the red mangroves throughout south Florida and the comparative abundance of the *L. angulifera* together with the large number of young spawned by each female leads one to believe that they would have been observed in plankton tows if they remain pelagic for very long. Lebour (1948) concludes that the veligers of *L. angulifera* are of the long planktotropic type but does not explain. Likewise, Thorson (1950) concludes from evidence presented in chart form that species which produce really large numbers of eggs have a long planktotrophic larval life with huge waste. It is evident that more research is necessary before a valid conclusion can be reached.

The veligers have been described by Lebour (1948). Measurements given by her for the Bermuda veligers were 120 to 140 μ . The measurements of veliger shells by the author have varied. An average of measurements in July, 1953, gave 92 μ for the length, 7.1 μ for the width, and 7.1 μ for the depth. The greatest length observed was 112.5 μ . The difference in size in the area under observation compared to the Bermuda veligers may be due to the warmer waters of the South Florida coast. The same phenomenon is exhibited in *Lacuna pallidula*, according to Thorson (1946, page 428). Larger eggs and embryos are found in Danish waters than off the coast of France. Also, from the same reference "*Brachystomia rissoides* has varying egg sizes in different localities even within neighboring areas".

A very large number of offspring is produced by each female during a single spawning period. Disregarding three cases where the spawn was less than a hundred, ninety percent of the individuals checked produced 10,000 to 100,000 offspring. Thirty percent were over 100,000. The actual counts, disregarding the three cases referred

to above, ranged from 300 to 760,000. The author found no indication of a correlation between the number of offspring produced and the size of the individual (Table 1).

TABLE I
SIZES OF FEMALES AND NUMBERS OF OFFSPRING PRODUCED.

Size mm	Number Checked	Maximum Number of Offspring	Minimum Number of Offspring
15	4	200,250	21,000
16	6	198,200	6,770
17	15	175,050	5,200
18	16	760,000	8,000
19	15	226,136	7,635
20	14	713,000	1,600
21	18	178,215	1,200
22	4	100,000	24,900
23	3	24,900	20,000
24	3	179,500	26,325
25	0		
26	3	239,000	3,500

The average number produced per individual also varies from one cycle to the next.

There does seem to be some relation between the amount of the spawn and the time of year. All individual spawnings of one hundred thousand or more (30% of those checked) occurred in March, April, the first spawning of May, or in September through December. No spawning of this size occurred through June, July, or August. These periods of the greatest individual spawning correspond in general to the periods where greatest percentages of females in the sample population were spawning. The large percentage of spawning females in June and July of 1953 did not produce the large numbers.

Growth rate and increasing maturity in L. angulifera. In a previous paper (1951) the author reported the observation that the growth rate of the female *L. angulifera* was greater than that of the male in the spring of the year. The snails used in the above mentioned report were returned to the mangroves on Biscayne Key until the following summer. Fifty-nine of the one hundred and ninety were recovered in June, 1952. Examination showed a similar differential growth rate of the two sexes extended throughout the year (see Figure 9). This same differential was found to be true in *L. littorea* by Moore (1936) and for *L. scabra* by Sewell (1924) as well as *Vivipara bengalensis*, a fresh water species observed by Sewell (1921).

Further study of the growth rate included observations of the

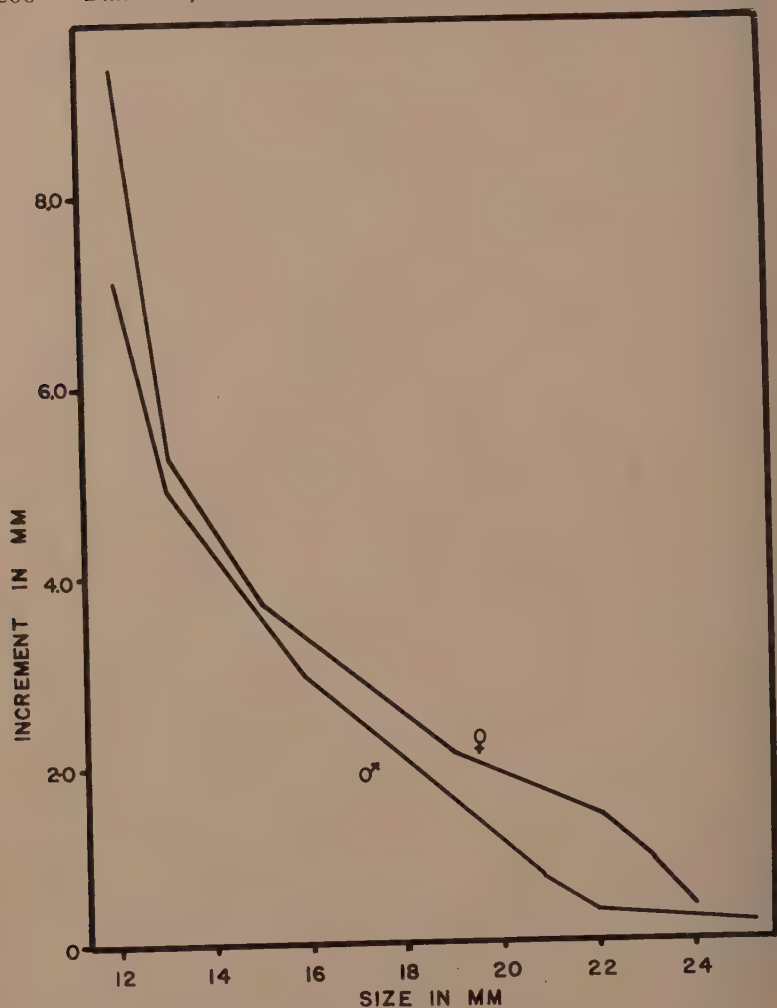


FIGURE 9. A comparison of shell increment of male and female in one year.

small snails (2.9 to 10.5 mm) for a six week period during two successive summers; a study of the growth of very small snails through the year from the summer, 1952, to the summer of 1953; a study of the growth made by mature snails from the summer, 1952, to the

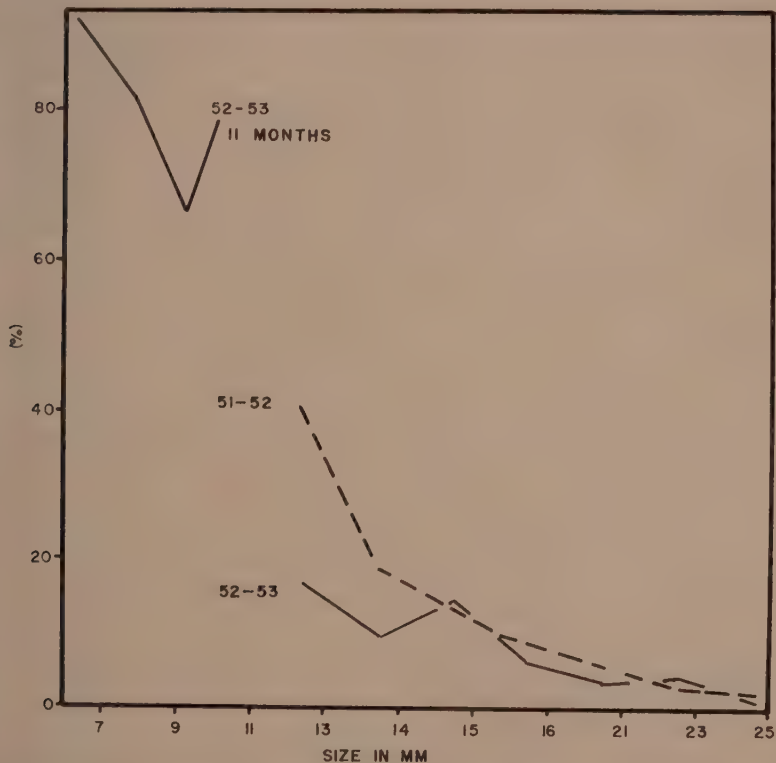


FIGURE 10. Percentage of increment of the original height of the shells in one year.

summer of 1953. The data for the latter are reduced to percentage increase of the original height and are plotted in Figure 10.

Work with the small snails was done in two different ways. In 1952, a population of 97 snails (3.9 to 8.6 mm) was divided into two groups with a similar size range. Since cellulose paint seems to be lethal to very young snails and notching results in severe shattering, the size distribution of the two groups was compared at the beginning and at the end of the time interval. It was found that (1), the largest increase in the large size was 35%; (2), the greatest increase in small sizes was 1.6%; and (3), the average increment of the twenty-seven largest snails was 18.6%, while the increment in

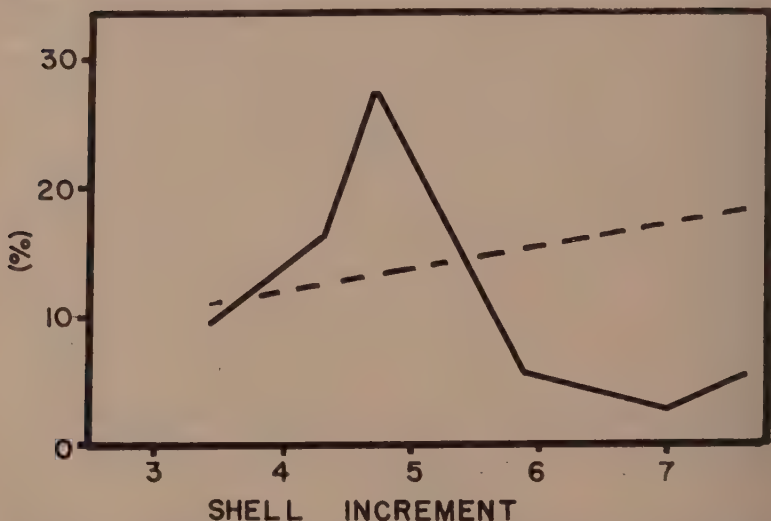


FIGURE 11. Shell increment in six weeks of the summer 1952 (broken lines) compared with that of the summer 1953 (solid lines).

the smallest snail was 11.6%.

In 1953 a population of 143 snails (2.9 to 10.8 mm) was divided into three size groups: A, those from 2.9 to 4.9 mm; B, those from 5.0 to 7.9 mm; C, those from 8.0 to 10.8 mm. Each group was placed on a mangrove root encased in a plastic screen. All twenty of those in group A were recovered in good condition; eighty percent of those in group B were in good condition; but fragmentation and death due to overcrowding and a more exposed location made it impossible to use group C in the analysis.

From Figure 11, it is evident that the very small snails grew at approximately the same rate in the two successive summers. Though the 1952 results are less precise it appears that the rate of growth for sizes 5 to 8 mm was about 13% greater in 1952.

The rate of growth of the immature snails viewed for the year and for the spring months show that on the whole the smaller snails grew faster than the mature ones and that the spring months may be the time when most of the growth is accomplished.

Since the very small sizes increase as much as 93% in a year, it seems that snails of 10 mm will be approximately one year old.

Since female snails of 11 mm have been known to spawn, it follows that maturity is reached during the second year. Sewell (1924) believed this was true for *L. scabra*, a species very similar to *L. angulifera*. Further, since snails of 10 mm increase approximately 50% per year, snails of 15 mm are about two years old. These increase approximately 15%, so snails of 17 mm are about 3 years old. Calculating thus from the data one sees that 19 mm snails are about 4 years old, 20 mm snails are about 5 years old.

Very few snails of more than 25 mm can be found on the north shore of Biscayne Key though *L. angulifera* of 35 mm have been collected from Ragged Key and other keys farther south. This may be due to a difference in growth rate but is more likely due to over collecting by tourists and the use of the snails in student projects at the University over the past five or more years.

The distribution of size in populations of L. angulifera. The data for population studies were contributed by Dr. Hilary B. Moore, Gilbert Voss, and Charles Gifford, as well as the author. Craig Phillips collected the last sample but measurements were made and distribution plotted by the author. The study continued from May, 1951, to August, 1953. Studies for the summer months are more complete than for the rest of the year. More data is desirable for the very small snails (2 to 10 mm).

Lack of data on the small sizes is due to the fact that collecting trips are usually scheduled for low tide. Unless it is a rainy or cloudy day the very young ones will not be out. These first year snails, undergoing the transition from an aquatic to a terrestrial existence, are apparently not so well adjusted to drying out as are the older ones. The latter can withstand drying conditions for five weeks (unpublished experiments by Bullis, Dow, Hutton, 1950; and Lenderking, 1951) and become active within twenty minutes after water, either salt or fresh, is supplied. In contrast, the author has found that the young ones have been killed in a very short time by the drying effects of acetone in cellulose paint during marking experiments, or by exposure to sun and heat even where there is splash, as on a sea wall. At low tide these young snails have been found 15 inches back in a dead mangrove root tangle. Even a quarter of an inch back under a cracked edge makes them difficult to see and collect. When the tide is rising these small ones move out of the cracks to the area of the root that is splashed and moist, preceding the tide up the roots. As the

tide recedes they move back down the root and into the cracks so that at low tide they are almost completely out of sight.

The roots of the red mangrove, *Rhizophora mangle*, provide an ideal arrangement for the young to migrate up and back with the tide without being exposed to excessive wind or sun. This seems to account for the greater abundance of the snails on the red mangrove rather than the black mangrove, *Avicennia nitida*, which grows in the same locations.

In spite of the fact that more complete data would be desirable, the available data can be correlated with the facts pertaining to growth and spawning given above. First, the greater part of each population illustrated is found to consist of individuals between the sizes of 15 and 25 mm, the greatest number of peaks falling between 17 and 20 mm. This correlates with the sharp decline in growth rate beginning with individuals of 15 mm. This would result in a continuous slow growth balanced by loss up to size 20 mm where there is a sudden dropping off of the population.

Spawning is probably extended but periodic throughout the year with the lowest rate in January and the greatest in April-May and October-November periods. If the size of one year old snails is about 10 mm and of the two year old snails is about 15 mm as shown above, there should be three, maybe four, small peaks between sizes 5 to 15 mm. Furthermore, these peaks should advance noticeably toward the big peak within a four to six months period with the peaks getting closer and merging between 15 and 17 mm.

The author believes the above expected results can be observed in the data as shown in Figure 12, which are a part, only, of the series obtained. For most of the sampling period the group under 15 mm in height is less clearly shown. A direct check of the growth against progression of peaks was made in the summer of 1952. In the population measured on June 20, a small peak appears at 8.0 mm. In the same population measured again on August 6, there is a comparable peak at 10.0 mm. In the growth studies conducted in this interval the average height of the largest individuals was 7.76 mm in June and 9.21 mm in August. This shows an increment of 18.6% over the original height in 36 days. This would move the peak from 8.0 mm to 10.0 mm if measurements are made only in whole millimeters. There is a similar progression from June, 1953, to August, 1953. Here the rate of growth was not as rapid but the population samples were measured after a longer interval, June 25 to August 31.

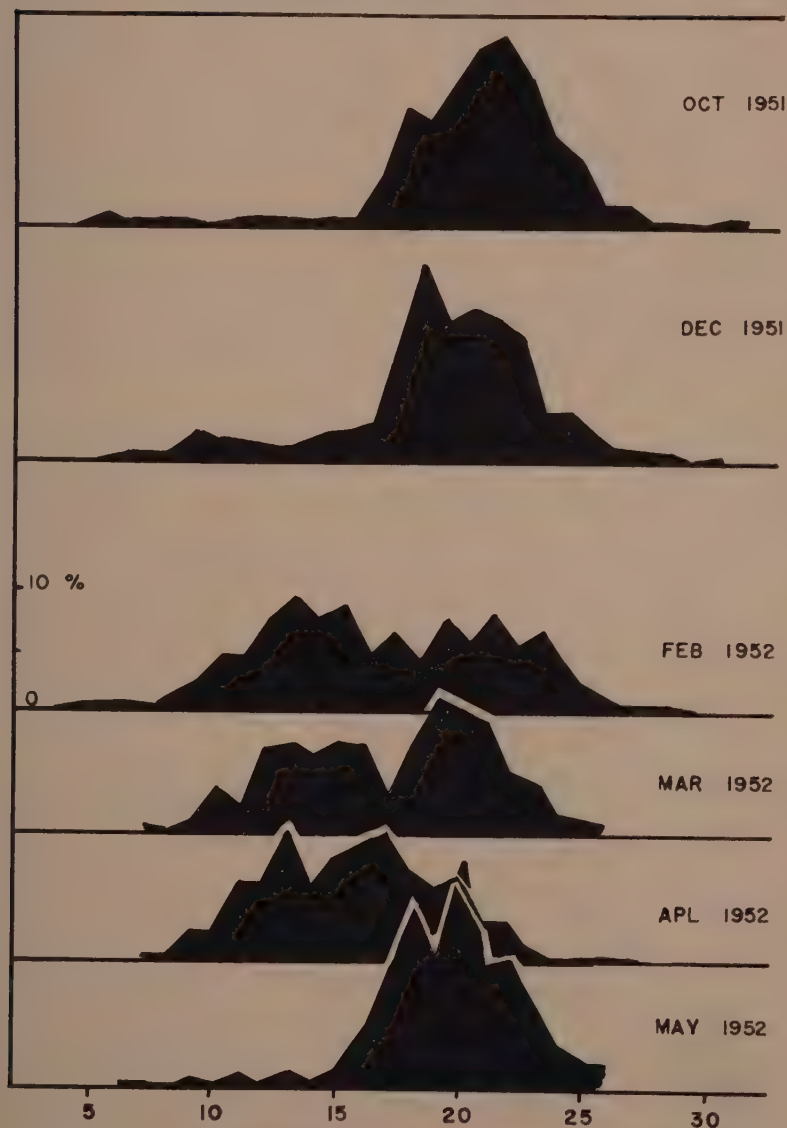


FIGURE 12. Population studies of *L. angulifera* from Biscayne and Virginia Keys.

DISCUSSION

In concluding, the data presented by the author will be compared to those of other observers on *L. angulifera* or related species.

The most recently published observations on *L. angulifera* other than those of the author were by Lebour (1948). She found that the *L. angulifera* in Bermuda spawned viviparously during July and August, and that the veligers were from 120 to 140 μ in length. Observations on the *L. angulifera* of Biscayne and Virginia Keys, by the author, indicate that spawning in this part of Florida occurs at least through ten months of the year and probably throughout the year, that the snails are oviparous and viviparous, shedding eggs in the early part of the cycle and veligers in the latter part of the cycle, and that the greatest length the veligers attained here in three weeks was 112.5 μ .

Sewell (1924) published work on *L. scabra*, an Indian species very similar to *L. angulifera*. The author found that the latter carries the eggs in the dorsal mantle cavity along the branchia, similar to the position described by Sewell for *L. scabra*. Those of *L. angulifera* appear as a bright yellow mass early in the cycle but may become dark later while Sewell describes the egg mass of *scabra* as brown. Sewell found *L. scabra* to be viviparous, the author found *L. angulifera* to be both oviparous and viviparous in that order in the spawning cycle.

Neither Lebour (1948) on *L. angulifera*, nor Sewell (1924) on *L. scabra* reported any indication of a lunar cycle. Lysaght (1941) observed that the planktonic egg-capsules of *L. neritoides*, a species living above high tide level as does *L. angulifera*, are shed only at high tide. The author has found that the spawning cycle of *L. angulifera* begins with shedding eggs sometime between the quarter phase of the moon and the new or full moon, and that by the time of new moon or full moon or soon thereafter, only veligers are shed. The peak occurs at this time and a diminishing number are shed until the succeeding quarter phase when there are none left in the mantle cavity. Thus a clear cut bilunar rhythm in spawning is evident, such as that suggested by Lysaght (1941) for *L. neritoides*.

Sewell (1924) found the greatest spawning in *L. scabra* occurred before and after the southwest monsoon. The present observer found peaks in spawning of *L. angulifera* occurred in April-May and the October-November periods. There is a probable correlation with rainfall in the Miami area.

Moore (1938) found that in *L. littorea*, females grow more rapidly than the males and that the younger snails grow more rapidly than older ones. Lenderking (1951) showed that in *L. angulifera*, females 15 mm or more in height grow more rapidly than males in the spring months. In the present paper it is shown that immature snails grow more rapidly than those over 15 mm in height. Growth studies extending through the year show that the rapid growth of the females compared to that of males is as indicated in the preliminary study during the spring months.

Both the varying rate of growth in the maturing snails and the spring and fall peaks of spawning are reflected in the size distribution of the population samples examined throughout these studies.

REFERENCES

- BULLIS, H., THOMAS DOW and ROBERT F. HUTTON
1950. An ecological survey of the family Littorinidae in South Florida. Unpublished report.
- CARSON, RACHEL L.
1951. *The Sea Around Us*. The Oxford University Press.
- FRETTER, VERA
1946. The genital ducts of *Theodoxus*, *Lamellaria*, *Trivia*, and a discussion of their evolution in Prosobranchs. *J. Mar. biol. Ass. U. K.*, 26: 312.
- GIFFORD, CHARLES
1952. Ecology of *Littorina angulifera*. Unpublished report.
- LEBOUR, MARIE V.
1935. The breeding of *L. neritoides*. *J. Mar. biol. Ass. U. K.*, 20 (2).
1945. Eggs and larvae of some Prosobranchs from Bermuda. *Proc. zool. Soc. Lond.* 25 (1).
1948. Eggs and larvae of the British Prosobranchs with special reference to those living in the plankton. *J. Mar. biol. Ass. U. K.* 22 (2): 105.
- LEIDY, JOSEPH
1845. Anatomical description of the animal of *Littorina angulifera*, Lam. *Phila. Junior Acad. Sci.* p. 344.
- LENDERKING, RUTH E.
1950. A study of the ecology of the *Littorina angulifera* of south Florida, particularly in the area of Bear Cut on Biscayne Key. Unpublished report.
1951. Observations on *Littorina angulifera* Lam. from Biscayne Key, Florida. *Quart. J. Fla Acad. Sci.* 14 (4).
- LYSAGHT, AVERIL M.
1941. The Biology and trematode parasites of the Gastropod *L. neritoides* L. on the Plymouth breakwater. *J. Mar. biol. Ass. U. K.* 25 (1).

MOORE, HILARY B.

1935. The biology of *Littorina littorea*. Part I. Growth of the shell and tissues, spawning, length of life, and mortality. Part II. Zonation in relation to other gastropods on stony and muddy shores. J. Mar. biol. Ass. U. K. 21 (2): 721.

MORRISON, JOSEPH P. E.

1950. Anatomy of the male genitalia of *Littorina angulifera*. Unpublished report.

SEWELL, R. B. SEYMOUR

1921. The banded pond snail of India, *Vivipara bengalensis*. Rec. Indian Mus. 22 (20) Part III: 215-292.
1924. Observations on growth in certain Mollusca and on changes correlated with growth in the radula of *Pyrozus palustris* (with a note on the radula by the late N. Annandale) Rec. Indian Mus. 26 (6): 529-548.

TATTERSALL, W. M.

1920. Notes on breeding habits and life history of the periwinkle. Dep't of Agri. and Tech. Inst. for Ireland Fisheries. Brit. Sci. Inves. 1: 1-11.

THORSON, GUNNAR

1946. Reproduction and larval development of Danish Marine bottom invertebrates, with special reference to the planktonic larvae in the Sound. Medd. Komm. Danm. Fisk.-og Havunders., sev. Plankton, 4. C.A. Reitzels Forlag-Bianco Lunos Bogtrykkovi Kobenhavn.
1950. Reproduction and larval ecology of marine bottom invertebrates. Biol. Rev. 25: 1-45.

WILSON, DOUGLAS P.

1951. Life of the shore and shallow sea. The McBride Co., New York.

A METHOD FOR RENDERING WOOD RESISTANT TO MARINE BORERS.¹

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ABSTRACT

The *in situ* precipitation of toxic materials in wood by double impregnation appears to have been inadequately tested as a borer-proofing method. A simple and economical method for borer-proofing wood by successive impregnations with copper sulfate and sodium hydroxide is described. This method has afforded complete protection to panels of pine and fir for more than two years. It appears to protect equally well against both shipworm and gribble.

INTRODUCTION

The replacement of wooden underwater structures damaged by marine borers is an expense faced by nearly every individual or group that maintains such structures. Damage from borers ranges from little or none in highly polluted waters to wide-spread practically complete destruction such as that which occurred in San Francisco Harbor in 1917-1921 (Atwood and Johnson 1924).

Most of the borer-proofing methods now in use involve the purchase of pre-treated lumber and timbers, or the expenditure of considerable sums for toxic paints or other surface-applied protective materials. The technique of borer-proofing by precipitation of borer-repellent chemicals *in situ* in the fibers of wood appears to have been largely overlooked. The only use of this technique seems to have been a successful double impregnation with copper sulfate and sodium carbonate tested by the Chemical Warfare Service (Atwood and Johnson 1924, pp. 207 and 214) which was borer-proof during the 161 days of test. Precipitation of toxin *in situ* in wood ensures penetration, avoids the search for specific solvents for toxins, avoids problems of adhesion failure, and should make available for testing almost every chemical compound which can be formed from the reaction of two solutions. The results presented in this paper are presented as an example of the economy and possible efficacy of the double impregnation method.

The double impregnation described here requires no specialized equipment, uses small quantities of inexpensive chemicals available

¹Contribution No. 680 from Woods Hole Oceanographic Institution.

at most drug stores, and uses methods available to everyone. It consists of precipitation of copper hydroxide in the fibers of the wood by soaking in copper sulfate and then in sodium hydroxide.² Wooden panels treated by this method have been afforded complete protection for more than two years in waters heavily infested with both gribbles and marine borers. This degree of protection appears to be comparable to the protection afforded by such related compounds as copper naphthenate, copper oleate, Cuprinol, etc.

METHODS

The impregnations consisted of soaking the wood for 48 hours in a saturated aqueous solution of technical grade copper sulfate, followed by a quick water rinse, and by 48 hours' soaking in an aqueous solution of technical grade sodium hydroxide (1/4 lb./gallon). Weights were employed to hold the wood submerged, and "sticks" to separate the boards and to hold them off the bottom of the bath. Both were shifted from time to time to enable the solutions to penetrate all the surfaces. At the end of the alkali impregnation the wood was rinsed repeatedly with water, and dried air-dry. After this it could be safely handled with the bare hands. Harmless white crystals of sodium sulfate usually appeared on the surface during drying.

Impregnation for 48 hours in each of the baths produced a dense layer of copper hydroxide 1/16 inch deep into the surface of the wood. Longer impregnation, of course, will produce deeper penetration. During immersion copper hydroxide appears to diffuse slowly inward into the wood, as well as outward to the sea water, for treated panels after immersion show a broad blue-stained band beneath the copper hydroxide layer (Figure 1).

Part of the panels were 3/4" x 6" white pine, part were 5/8" x 4" fir, all were one foot long. The test pairs were hung from a horizontal brass chain 2-3 feet below lowest low water. The panels were assembled in pairs, each treated panel being paired with an untreated control panel which had been immediately adjacent to it in the original board. The suspending chain made a round turn about the brass bolt which held the pair together at their upper ends; the turn of chain served as a spacer between the panels. The lower ends of the panel pairs were bolted together in the same fashion as the upper ends. The treated panels were put at the head ends of the bolts so that they

²A recent report (Baechler, 1953) describes a double impregnation method which uses copper sulfate and sodium chromate in the preservation of fence-posts.

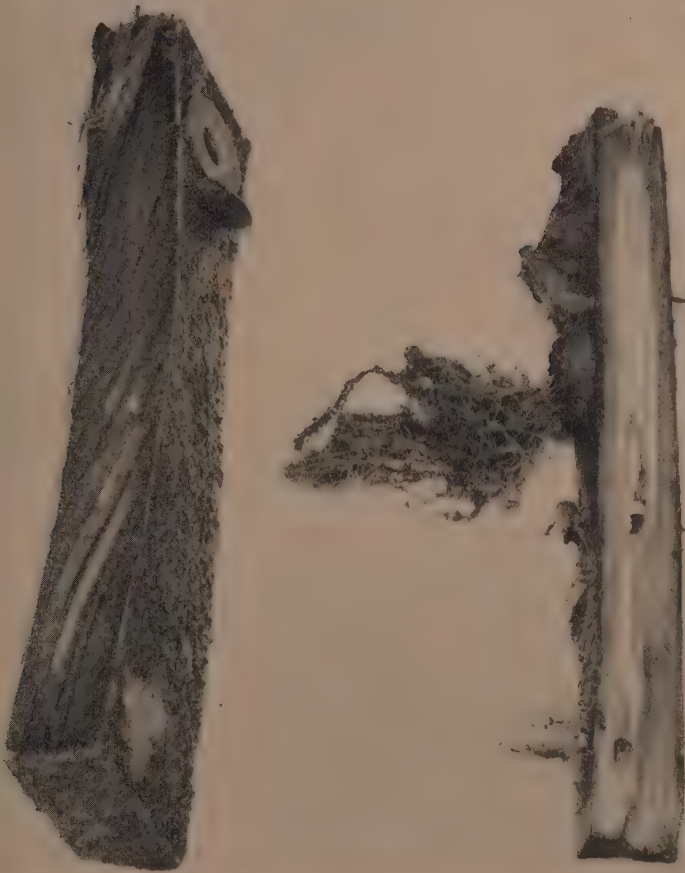


FIGURE 1. Treated panel (left) and untreated control panel (right) of pine after 13 months' exposure beneath the Woods Hole Oceanographic Institution dock, Great Harbor, Woods Hole, Mass. The layer of basic copper carbonate in and on the surface of the wood of the treated panel shows as a light band; the dark color of the inner wood of the treated panel is due to the copper hydroxide which appears to diffuse inward as well as outward during submergence.

could be identified at all times. The identifying number of each pair was stamped into the bolthead at the top of each pair.

RESULTS

The first set of test panels was suspended beneath the Woods Hole Oceanographic Institution's pier. Here it survived 1½ years before it was carried away during its second winter. One pair of panels was removed after 13 months' exposure. At that time the treated panel was moderately fouled but was free of borers, while the control was heavily fouled and contained two large borers (Figure 1). This test was not considered significant.

A second set of panels, 5 pairs in all, were hung from the Marine Biological Laboratory's floating dock in the Eel Pond at Woods Hole, Mass. Here they remained from 22 April 1949 until 8 September 1951. The waters of the Eel Pond are shallow, warm, and heavily infested with borers.

After 14 months in the Eel Pond the control panels were becoming narrow as borer and gribble (*Limnoria*) damage allowed their edges to erode away. They were honeycombed with borer and gribble tunnels and fairly heavily fouled with hydroids, while the treated panels were free from fouling and showed no visible damage of any kind.

By 8 September 1951 the control panels had disappeared except for small areas of badly riddled wood immediately around the washers and bolts. The conditions of the panels as removed from the water are shown in Figures 2 and 3. Each treated panel was split for internal inspection. Not a single borer was found in any of the treated panels, no gribble damage was present, nor (with one exception) were their surfaces more than very lightly fouled with hydroids and bryozoa (Figure 4).

DISCUSSION

At the termination of the test the treated panels were verdigris green in color and somewhat slippery to the hand. The green color was due to basic copper carbonate formed on and in the wood surface as copper ions leached slowly to the surface and came into contact with the oxygenated and carbonate-rich sea water. The basic copper carbonate is apparently either toxic or distasteful to gribbles and the larvae of borers.

The copper hydroxide precipitated in the wood is only very slightly soluble and consequently can supply copper ions at a slow rate for a

long period of time. After two years and four months of exposure the copper hydroxide layers in the treated panels were not visibly decreased in thickness nor appreciably dulled in color. They apparently contained reserves of copper adequate for still longer exposure.

Cost estimates

The 25 square feet of lumber used in the tests and preliminary trials were treated with 5 lbs. of copper sulfate (more than 4 pounds of which were recovered by allowing the sulfate bath to evaporate after the test panels were prepared), and 1 lb. of sodium hydroxide. The total cost of chemicals for the treatment was about \$1.90. In practical application, the construction of two water-tight copper-fastened wooden troughs for the solutions would be a further, but usually not excessive, cost. The troughs need to be built only slightly longer, wider, and deeper than the wood to be treated.

Lumber or timbers to be treated by this method should be pre-cut to size so that the end-grain as well as the sides may be penetrated and protected. If the weights atop the lumber and the "sticks" beneath and among it in the soaking troughs are shifted occasionally both the sides and ends will be treated simultaneously and evenly, giving uniform protection.

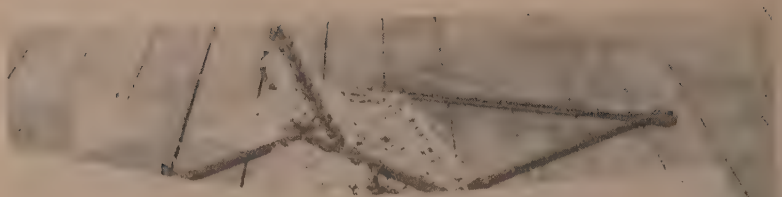
The method is best applied to pre-cut lumber and timbers, though untreated wood exposed by carpentry can be treated by several successive swab applications of the sulfate followed by several swabbings with caustic.

This method offers little promise in the field of piling protection where pressure-impregnation with creosote is general practice. The toxicity imparted by this method appears to have definite long-term protection value, but it is in the surface of the wood and this makes it less suitable for structures subject to heavy battering by boats, ice, flotsam, etc., for small areas of untreated wood bared by battering would allow borers to penetrate the untreated interior wood. Structures subject to less severe battering may be protected by treated bumper strakes and corner-trim applied over treated timber.

Cautions

Copper sulfate is an effective skin poison and should not be left in contact with the skin for more than a few minutes.

Sodium hydroxide is a strong caustic and should be kept from contact with the skin, mouth and eyes. Loops of copper wire, rubber hose, rope, or other non-ferrous material should be used to remove wood from the caustic bath.



2



3



4

The copper salts in and on the wood greatly accelerate the corrosion of iron. Any structures fabricated of wood so treated must be brass or copper-fastened. Iron in contact with the treated wood will not only corrode with excessive rapidity, it will inactivate the copper and prevent its beneficial effects.

Caustics other than sodium hydroxide probably would work equally well. In the absence of sodium hydroxide, ordinary lye could be used. Ammonia will *not* substitute as the caustic, for it unites with copper to form a *soluble* complex hydroxide which will dissolve out of wood.

SUMMARY

Double impregnation affords a means of precipitating *in situ* in the fibers of wood any toxic material which can be formed by the reaction of two solutions. An efficient and economical example of borer-proofing by double impregnation is described.

The method consists of surface impregnation of the wood with copper sulfate, followed by a sodium hydroxide bath which precipitates copper hydroxide in the wood. Exposure to sea water produces a toxic layer of basic copper carbonate on and in the wood surface.

Woods panel treated by this method have been afforded complete protection from marine borers and from gribbles (*Limnoria*) for

FIGURE 2. Outer surfaces of the treated panels after two years and four months' exposure beneath the floating dock of the Marine Biological Laboratory, Eel Pond, Woods Hole, Mass. The panel at the left was fouled rather heavily with hydroids for reasons which are not known. The notch in the lower edge of the panel on the right is part of an auger hole which was present in the original board.

FIGURE 3. Inner surfaces of the treated panels after two years and four months' exposure in the Eel Pond, Woods Hole, Mass. The borer-riddled remains of the untreated control pane's may be seen atop each panel. A few tube worms and barnacles occurred on these inner surfaces of the treated panels.

FIGURE 4. The treated panels, arranged in the same order as in Figure 3, after being split through the lower bolt-holes and turned to expose the split edges. A few strands of hydroid lie across the split edges of the panel on the left. The lower bolt-hole shows in the lower end of one or both parts of each panel. The lower central portion of the panel on the right shows a nail-hole which was present at the time the panel was prepared. No borers were present in any of the treated panels. The lower bolt from each panel pair, with its fragment of the lower end of the control panel has been laid out beneath each treated panel. The condition of the upper fragment of the control panel shows well at the left of the upper end of the panel on the right.

more than two years, while untreated panels of identical wood were completely destroyed.

The cost of chemicals, treatment troughs, etc., is less than the cost of commercial antifouling paint.

The chemicals required are available at most drug stores.

The copper sulfate and sodium hydroxide may be of the crudest technical grade.

The chemicals used are poisonous and must not be allowed to remain in contact with the skin, nor should they be allowed to enter the mouth or eyes. Treated wood after being washed and dried can be safely handled.

The treated wood requires brass or copper fastenings. Iron in contact with it destroys the protective value of the copper salts and is very rapidly corroded away.

REFERENCES

ATWOOD, W. G. AND A. A. JOHNSON

1924. Marine structures, their deterioration and preservation. National Research Council, Div. Eng. and Indust. Res., Committee on Marine Piling Investigations, Washington, D. C., 534 pp.

BAECHLER, ROY

1953. How to treat fence posts by double diffusion. Report No. 1955, U.S. Dept. of Agr., Forest Service, Forest Products Laboratory, Madison, Wisconsin.

A NOTE ON THE SHEDDING OF PIGMENT FROM THE BODY WALL OF *HOLOTHURIA GRISEA* SELENKA.

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The conspicuous colours of many echinoderms have been attributed to the accumulation in the body wall of supposedly excretory substances, some of which have been said to leave the body (Lison, 1930; Cuénot, 1948). Their metabolism is not understood and some have not been chemically identified. In the holothurians in particular, transport of the pigment has been attributed to amoebocytes, (Cornil, Mosinger and Calen, 1935; Millott, 1953) which, Cuénot states, eliminate the pigment by diapedesis, passing through the wall of the respiratory trees, intestine, polian vesicles, etc. Though experiments show that this can occur in the body wall as well as in these organs (Millott, 1950), there is little or no direct evidence concerning the quantities of naturally occurring pigments that are eliminated in this way. Indeed, some maintain that the pigment accumulates with age (Clark, 1907). A direct observation of the shedding of a considerable quantity of pigment from the body wall of *Holothuria grisea*, is therefore worth recording.

One of three individuals kept in an aquarium, was seen to assume a ragged appearance due to the loosening of what appeared to be the superficial body wall. During the ensuing 48 hours, the loosened portion became free and eventually rolled up, forming a more or less complete cast, heavily charged with pigment. In it the outline of tube feet could be recognised.

At first, it was thought to be pigment laden mucus, but on examination it appeared tougher and could be teased out on a slide to form a chocolate coloured film. When this was examined under a microscope, it was seen to be an exact replica of the surface of the holothurian, with projecting tube feet, each complete with a cast of the terminal sucker. In many cases, the cast of the tube feet was turned inside out, that of the sucker being introverted within the tube, indicating that the cast had been loosened from the general body surface first and then drawn off from the tube feet. It was densely suffused with pigment granules, most being deep brown, a few, yellow, but no cellular structure was discernible, possibly owing to the dense accumulation of pigment.

The process thus appeared to represent the casting off of pigment laden skin, rather than of mucus.

The solubility of the pigment was examined, portions of the cast being extracted with both hot and cold solvents for 12 hours. In the case of the hot solvents, extraction was performed beneath a reflux condenser. The yellow pigment dissolved out rapidly in both cold and hot ethanol. The brown proved much more resistant, being insoluble in water, 90% ethanol, acetone, benzene, petrol ether, diethyl ether and chloroform. It dissolved in ethylene chlorohydrin and slightly in pyridine and 1.0 M HCl.

Since no more casts were produced, no more pigment was available for further tests, but it is clear that in its solubility, the pigment resembles melanin (Lea, 1945), and the melanin-like pigment of *Holothuria forskali* delle Chiaje (Millott, 1953).

The process was not seen to occur again in this specimen nor to take place in any of the other individuals. It is as yet impossible to be sure that it was normal, but it is significant that the individual which cast off the pigment lived as long as the others, leading an apparently normal life for several weeks after the event.

Only continued scrutiny of such forms will provide the answer. It is desired to direct the attention of observers to this process, since it may well have considerable significance in relation to the metabolism of pigments, especially those of the melanin type, about which we have little knowledge.

REFERENCES

- CLARK, H. L.
1907. The Apodous Holothurians. *Smithson. Contr. Knowl.* 35: 64.
- CORNIL, L., M. MOSINGER AND M. CALEN
1935. La désintégration physiologique de l'appareil pigmentaire chez les holothuries. *C. R. Soc. Biol., Paris.* 119: 106-7.
- CUÉNOT, L.
1948. *Traité de Zoologie* vol. 11. P. Grassé, Masson, Paris.
- LEA, A. J.
1945. A Neutral Solvent for Melanin. *Nature, Lond.* 156: 478.
- LISON, L.
1930. Recherches histophysiologiques sur les amibocytes des echinodermes. *Arch. Biol., Paris,* 40: 175-203.
- MILLOTT, N.
1950. Integumentary pigmentation and the coelomic fluid of *Thyone briareus* (Lesueur) *Biol. Bull., Woods Hole,* 99: 343-4.
1953. Observations on the skin pigment and amoebocytes, and the occurrence of phenolases in the coelomic fluid of *Holothuria forskali* delle Chiaje. *J. Mar. biol. Ass. U. K.* 31: 529-539.

MARINE FUNGI IN BISCAYNE BAY, FLORIDA.

II. FURTHER STUDIES OF OCCURRENCE AND DISTRIBUTION¹

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ABSTRACT

A survey in Biscayne Bay, Florida, of the marine fungal population, reveals the presence of seven morphologically different halophilic wood-inhabiting fungi, all of the general class Ascomycetes. Five of these fungi are described here, while two forms have been described in the initial paper of this investigation. All of the fungi are true marine forms, occurring consistently on wood submerged in marine areas. Forms similar to fungal types found in Biscayne Bay, have been discovered in investigations at Bimini, Bahamas, across the Gulf Stream from the Florida coast. Studies over a period of 18 months indicate noticeable variations in concentrations of the fungi coincident with differences in the biological activity of the area.

The fungi described are studied in pure culture on salt water agar, tabulating mycelial growth rates and the mode and rate of ascocarpic production. Correlated cultural studies and field collections show a high rate of fungal activity, relative to the different fungi, thus enabling the comparison of the biological processes of the individual fungi on wood submerged in marine environments.

INTRODUCTION

In the initial paper of this series on marine fungi in subtropical waters (Meyers, 1953), it was shown that, in general, these halophilic fungi comprise a regular and significant part of the microfloral population of this area, particularly that occurring on submerged wood. The introductory survey dealt with two different and morphologically variable marine fungal forms and, within the scope of the study, tabulated their occurrence and relative distribution in Biscayne Bay, Florida. Since these studies, five additional marine fungi have been isolated from submerged wood, and their description, occurrence and distribution, as well as a total representation of fungal density in Biscayne Bay, is presented currently.

To facilitate the collection of as many wood-inhabiting marine fungi as possible and to study the effects of submergence periods of more than 6 months on the fungal complex, additional stations, together with those listed in the previous survey, have been established.

¹Contribution No. 117 from the Marine Laboratory, University of Miami. This study was aided by a contract Nonr 840(01) between the Office of Naval Research, Department of the Navy, and the University of Miami.

Wherever possible, the incidence of fungal infestation at the different stations, particularly that of Form No. 2, has been presented in conjunction with observed accompanying environmental factors of the immediate wood substrate and adjacent marine area.

In order to define more accurately the growth characteristics of the individual fungi as observed on the wood surfaces, the fungi in this survey all have been isolated in pure culture on salt water agar and their growth studied. The adaptability of these halophilic organisms to artificial cultural techniques permits the analysis of approximate rate of development, both mycelial and ascocarpic, and the correlation of these observations with those of the survey.

An investigation also has been made of the marine fungal population at Bimini, Bahamas. The location of Bimini at the eastern edge of the Gulf Stream permits the study of factors of fungal occurrence and concentration as correlated with ecological conditions different from those in Biscayne Bay.

As part of the distributional studies of the marine fungi, wood panels have been submerged at other localities in the Bahamas and Caribbean as well as in the Gulf of Mexico and along the Atlantic seaboard. While some mention is made here of the occurrence of fungal forms in certain of these areas, the complete results from all of the localities will be given in a later publication. The examination of marine fungi over such a wide area affords excellent means of correlating diverse morphological fungal characteristics and ecological differences of the various marine areas with fundamental physiological studies of the individual fungi concerned.

The writer wishes to express his sincere appreciation to Dr. E. S. Reynolds of the Marine Laboratory, University of Miami, for his invaluable suggestions and guidance throughout the entire investigation. The studies at Bimini, Bahamas, were made possible through the cooperation of Dr. C. M. Breder, Jr. of the American Museum of Natural History in the use of the facilities of the Lerner Marine Laboratory at Bimini. The hospitality and cooperation of Dr. and Mrs. M. B. Bishop of the Lerner Marine Laboratory is sincerely appreciated.

METHODS

The marine fungi described were collected on yellow pine and northern spruce panels, using the methods given in the earlier survey. This arrangement of the wood in a "sandwich-like" manner was found

to be particularly advantageous, for the predominant ascocarp population was consistently evident on the two inner wood surfaces separated by the width of the supporting chain. The use of northern spruce panels allowed the study of the possible effect of two different wood substrates upon the diversity and intensity of fungal attachment. This analysis of fungal infestation of woods of different anatomical structure is presented in the discussion on fungal occurrence and distribution. The panels were submerged continually from 9 weeks to a year, varying at the different stations according to the degree of biological destruction of the wood.

The stations (Table 1) at which panels were submerged were established so as to include diverse ecological habitats where different fungi, concentrations, and relative environmental factors could be studied. Stations No. 1 and Nos. 4-10 are those listed previously (Meyers, 1953), while Stations No. 2 and 3 and 11-15 represent additional areas where panels were submerged. The area of Stations No. 1-9 represents a part of the Bay where moderate to high amounts of sewage products occur, together with an enhanced rate of growth of fouling organisms (Smith, et al., 1950). Stations No. 10-15, near Key Biscayne and in the southern Bay area, are not adjacent to the built-up dock and city areas and also receive considerably less direct sewage flow. The location of the different stations is shown in Figure 1.

In isolating the fungi from the natural substrate, it first was necessary to remove the excess surface moisture from the panels by wrapping the wood in absorbent paper, followed by drying in an incubator at 32-36°C. for 1-2 days. The drying process resulted in the exudation of ascospores from the fruiting structures of the fungi on the surface of the wood. Thus, it was possible to transfer these ascosporic masses, relatively free from bacterial contamination, by a sterile needle to salt water agar plates. Repeated transfers of succeeding generations of vegetative and reproductive inocula enabled the establishment of pure cultures of the different fungi.

The medium used for the study of growth characteristics of the individual fungi was two percent Bacto agar made up in filtered aged (ZoBell, 1946) sea water with no added nutrients. All of the fungal colonies were established in the early isolation work by single ascospore inoculation to insure purity of culture and comparative maturity of growth. The procedure followed in studying the radial growth rate of the fungi in culture was the use of small blocks of inoculum, 1-3 millimeters square, dissected from the individual stock cultures. The



FIGURE 1. Map of Biscayne Bay showing location of stations.

inoculum usually was taken from the new mycelial growth around the periphery of the colony. Radial growth rate was determined by measuring the spread of the mycelium from the agar block inoculum onto the new salt water agar layer. Following inoculation, the colonies were permitted to grow for 3-4 days before the initial measurement so as to facilitate the hyphal establishment on the agar base. Measurements were made radially from the four edges of the agar block inocula to the outer margin of mycelial development. After 6-8 days, the colonies were measured again, and the rate of radial growth was taken as the increase in radius of the colony in millimeters per day between the first and second measurements. The value for any one measurement was taken by averaging approximately 20 readings or colonies used for each of the fungi. In addition to colonies established by mycelial transfer, studies were made of the fungal growth from single spore transfers of certain of the fungi studied.

Viable pure cultures of the fungi described, and prepared slides of the different perithecia mounted in lactophenol and sealed with a phenolic resin (Tufon No. 74), have been preserved.

TABLE 1

STATIONS ESTABLISHED FOR STUDY—SEE FIGURE 1

- | | |
|----------|--|
| (No. 1) | Fisher Island near Government Cut |
| (No. 2) | City Terminal Yacht Basin |
| (No. 3) | Marker No. 3, Miami Beach side of Government Cut |
| (No. 4) | Flamingo Hotel docks |
| (No. 5) | Beach Boat Slips |
| (No. 6) | Marker No. 22 |
| (No. 7) | Marker No. 16, adjacent to Miami dock area |
| (No. 8) | Marker No. 14, near mouth of Miami River |
| (No. 9) | Florida East Coast Railroad bridge, within Miami R. area |
| (No. 10) | Virginia Key, southwest tip |
| (No. 11) | Marker No. 28, off Key Biscayne |
| (No. 12) | Hurricane Harbor, Key Biscayne |
| (No. 13) | Cape Florida Marker 2 x, southeast corner Key Biscayne |
| (No. 14) | Dinner Key Yacht Basin |
| (No. 15) | Matheson Hammock Boat Basin |

DESCRIPTION OF FUNGI

The following descriptions of the various morphological features of the fungi collected in Biscayne Bay are the compilation of both cultural characteristics and features of the organisms as they appear on the natural substrate. The ascospore size and appendiculate configuration are described following the examination of a wide range of ascocarps of the particular fungus, from different panels and stations as well as various pure cultures. The different fungi collected

are designated in this paper as "forms" to facilitate further classification of species. The variability of the ascocarpic character within the forms currently described necessitates additional study of this fungal structure before the limitations of the species can be established.

Certain diagnostic structural features apparently serve to adjust these fungi to the aquatic environment. These include the early breakdown of the ascus wall very shortly after the ascospores have been delimited, thus facilitating the dispersal of the ascospores singly into the aquatic medium, and the distinctive appendiculate condition of the ascospores, which may serve to facilitate orientation in the water and attachment to submerged surfaces (Figure 2, A-H).

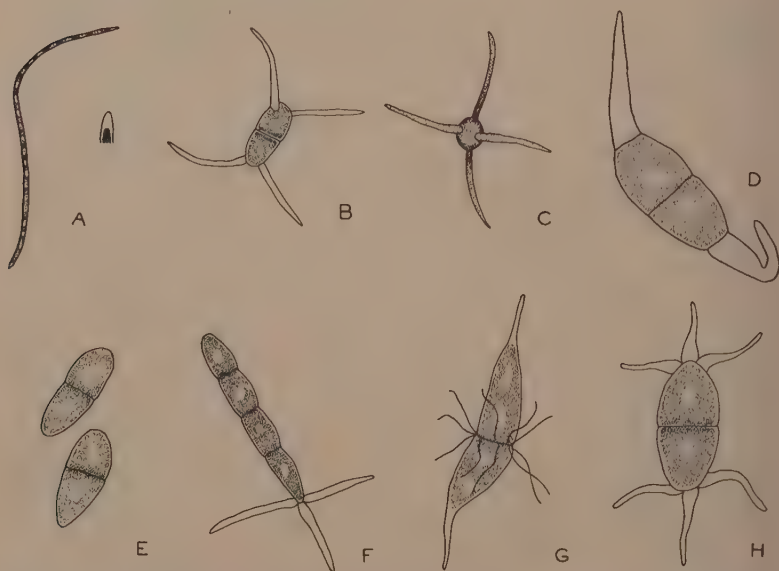


FIGURE 2. A, Characteristic filamentous *Halophiobolus* (Form 1) ascospore. Inset shows enlargement of hyaline terminal appendage. Ascospore approximately 115 times actual size. B, Bi-appendaged 2-celled ascospore of Form 2, X approx. 330. C, Form 2 spore showing end view with arrangement of appendages at the spore apices, X approx. 330. D, Form 3 ascospore with single stout appendage at each apex, X approx. 660. E, Form 4, 2-celled ascospores, X approx. 500. F, Ascospore of Form 5 showing appendage arrangement at terminal end, X approx. 660. G, Form 6 ascospore with single appendage at each apex and cilium-like appendages around middle septum, X approx. 830. H, Form 7 ascospore showing tri-appendiculate arrangement at each apex, X approx. 800.

Forms No. 1 and 2 have been described previously (Meyers, 1953) and Forms No. 3 and 6 apparently are similar to genera described by Linder (1944). The latter two are described for the first time from a subtropical marine environment, since the previous description of the genera was from the New England area. Forms No. 4, 5 and 7 do not appear to have been reported previously in mycological literature.

FORM NO. 3

Ascocarps: (as produced on the wood panels) slightly to fully imbedded, with exserted neck, gregarious; dark fuscous when mature, membranous and therefore pliable walls, globose venter, ostiolate, 400-500 (including neck) x 164-200 microns; neck central, light fuscous, 250-300 x 19-24 microns, pubescent-like hyphal growth from periphery of neck wall, becoming fimbriate at neck apex.

Asci: hyaline, becoming deliquescent on maturity.

Ascospores: 2-celled, hyaline, 23-25 x 7.8-8.5 microns, elongate-ellipsoid with a single stout hyaline tapering straight or curved appendage at each end; appendages 23-39 microns long, at one end long and acuminate, at the opposite end shorter and obtuse, occasionally deciduous leaving the spore with truncate apices, slight indentation in basal truncate portion of appendage (Figure 2, D).

The Form No. 3 fungus appears to be similar to the genus *Ceriosporopsis* Linder as indicated by the appendiculate nature of the ascospores, the deliquescent asci and deciduous appendages, and the similarity in size and general description of the ascospores as given by Linder. However, the perithecia isolated from the natural substrate and in artificial culture differ considerably in morphological characteristics from those described by Linder as being typical of the genus. The latter are "ovoid to elongate-ellipsoid, ostiolate; the ostiole eccentric, rounded-conoid or papilliform." The perithecia isolated from the wood substrate and also as developed in pure cultures in this study have, in general, a definitely globose base and a conspicuously elongated neck which is not eccentrically placed.

In addition, this fungus shows great variability in artificial culture in both number of necks occurring on individual ascocarps and degree of modification or distortion of the neck. This morphological variability is the subject of a paper currently in preparation, in which the matter of variability and taxonomic position is discussed. Form No. 3, in general, differed from the other fungal forms in a decidedly

greater range and intensity of ascocarp variation.

FORM No. 4

Ascocarps: superficial to deeply imbedded with exserted neck, gregarious, black, carbonaceous, globose to subglobose venter with basal portion often flat, 500-700 (including neck) x 270-320 microns, ostiolate; neck mostly terminal, straight to curved, black at base becoming sub-hyaline to hyaline toward apex, neck wall well defined, nonstriated, 370-428 x 38-42 microns, occasional slight bulbous swelling at base.

Asci: 8-spored, clavate, rounded apex with slightly tapering base.

Ascospores: 2-celled, non-appendaged, ellipsoid (rounded apices), 21-23 x 7-8 microns (Figure 2, E).

While the ascospores of this form are apparently non-appendaged, the consistent isolation of the fungus from wood continuously submerged in salt water, as well as the growth and *reproduction* of the fungus on salt water agar, establishes the organism as a normal halophilic form.

FORM No. 5

Ascocarps: (as produced in salt water agar culture) imbedded with neck exserted beyond substrate, scattered, sub-hyaline below and dark fuscous above, becoming dark brown to black at maturity, tangled slightly spreading layer or hyphal mat around upper portion of ascocarp, globose, ostiolate, 570-600 (including neck) x 310-350 microns; neck central, approximately 32 microns in diameter, sub-hyaline, pubescent-like growth along neck wall formed by extension of hyphae comprising outer neck periphery.

Asci: not seen.

Ascospores: 4-celled, appendiculate, elongate-cylindrical, rounded apices with slight constriction at septa, sub-hyaline, 31-39(35) x 4-5.5 microns, basal and apical cells smaller than middle cells, breaking readily at the septa into mono- and dicellular segments; appendages, 3, arising from one end, deciduous or deliquescent, hyaline, flexible, acuminate, 19-23 microns long, approximately 2.5-3.0 microns in diameter at base; one appendage inserted at end of the spore, with remaining two appendages slightly behind the spore apex, arranged at 90° to the terminal appendage and 180° to one another (Figure 2, F).

The taxonomic features of this form are described from the structures produced in artificial culture, owing to the relative scarcity of

ascocarps of the fungus on the wood substrate. However, the appearance of the ascocarps on the salt water agar was similar to the type of ascocarps found on the exposed wood.

FORM No. 6

Ascomycetes: solitary or scattered, often superficial on calcareous deposits on wood surface, spherical, 245-280 x 190-245 microns in diameter, carbonaceous, black with a metallic luster, ostiolate; ostiole small and papillate, apical.

Asci: fusoid or broadly fusoid, rounded to slightly tapering apex, 65-80 x 16-20 microns.

Ascospores: hyaline, 8, fusoid (at times slightly curved), biappendiculate, 2-celled with slight constriction of the spore wall at the septum, several small globules in each cell, 22-29 (excluding the appendages) x 6-8 microns, single hyaline elongate subcylindrical appendage with obtuse apices at each end, and numerous (between 10-20) cilium-like appendages immediately above and below the central septum; terminal appendages 9-13 microns long and approximately 1 micron in diameter at the base of the spore; cilium-like appendages about half the length of the spore, occasionally deciduous (Figure 2, G).

This Form No. 6 fungus appears to belong to the genus *Peritrichospora* Linder, as indicated by the numerous cilium-like appendages around the central septum and by the carbonaceous spherical perithecia with proportionately very small ostioles.

FORM No. 7

Ascomycetes: imbedded within wood tissue, sub-globose to elongate, apices obtuse, black, carbonaceous, occasionally ridged (probably by delimitation of surrounding wood tissue), 300-425(350) x 210-280 microns, ostiole very small, slightly papillate or merely an opening in the ascocarp wall.

Asci: not seen but probably 8-spored since the ascospores occur frequently in bundles or clumps of eight.

Ascospores: ovoid, hyaline, appendiculate, rounded apices, 2-celled, 19-23 x 8.1-11 microns; appendages hyaline, 6-8, acuminate, occasionally curved, 12-16 microns long, 1.5-2.3 microns wide at the basal portion; 3-4 appendages at each apex, arranged fascicle-like around spore tip. Certain appendages at times deciduous, leaving spore biappendaged at both apices (Figure 2, H).

GROWTH FEATURES OF THE FUNGI

Ascospore Germination: The ascospores of Forms No. 1-7, from asco-

carps on the wood substrate and in artificial culture, germinated within 18 to 48 hours on a salt water agar base. The optimum time of ascospore germination and the rate of growth of the germination hyphae for the initial 48 hour period are given in Table 2. The viability of the ascospores of the fungi was evidenced by the large percent of visible spore germination, even from ascocarps produced on cultures 5-6 months on salt water agar. In some plates containing several hundred ascospores of an individual fungus, germination was apparent in over 75 percent of those spores examined.

The rate of elongation of the germination hyphae, as well as the time of germination of the ascospores, varies among these different fungi. The most vigorous radial growth was apparent in the germinating ascospores of Form No. 1, also characterized by the shortest period necessary for optimum spore germination. While the growth rate of Form No. 1 was equally rapid during the initial germination period and after the establishment of the fungal colony, the other fungi showed a decrease in radial growth following early germ tube elongation. This was noticeable particularly in the case of Form No. 7 where a rapid early rate of mycelial spread contrasted with a much slower later growth rate.

The several-celled ascospores of Forms No. 1 and 5 were characterized by germ tube production from two or more cells, often with vigorous branching of the primary hyphae. The mode of germination of the 2-celled ascospores of Forms No. 2, 3, 4, 6, and 7 was usually from one apex, although germination from both apices was apparent in some forms, particularly Form No. 6. In addition to number of primary germ tubes, the fungi differed in amount and rapidity of branching during the initial 24 hours following spore germination.

TABLE 2
RADIAL GROWTH ON SALT WATER AGAR

Form	Initial Ascospore Germination	Germ Tube Elongation (48 hrs.)	Mycelial Growth in mm. per day
No. 1	<18 hrs.	1200-1400 microns	2.9
2	<48 hrs.	128- 214 microns	0.5
3	<48 hrs.	*	0.86 (0.8-1.1)
4	<48 hrs.	*	<0.30
5	<48 hrs.	250- 420 microns	0.71
6	<48 hrs.	500- 780 microns	2.1 (1.8-2.3)
7	18-24 hrs.	710- 780 microns	<0.30

* Not recorded during current experiment.

Mycelial Development: Although the ascocarpic stage is recognized quite readily on the exposed wood, the mycelial stage in the submerged wood can be ascertained only by careful sectioning and microscopic examination of the wood. Hence, while the presence of ascocarps establishes the occurrence of the accompanying fungal mycelium in the wood, cultural studies are necessary to study the mycelial developmental rates of the individual fungi. The millimeters of radial growth per day coupled with the intensity of ascocarpic formation on the agar medium and on the wood substrate, establishes an index for evaluating the relative vigor of the different fungi throughout the infection period.

The rate of mycelial development of the various marine fungi is given in Table 2. The observed increase in colony size for the individual fungi is based on actual radial growth in the agar medium and excludes, to some degree, differences in density of the fungal colonies. While variations occurred between the fungi in hyphal branching and aerial growth, it was only in Form No. 1 that the development pattern differed considerably from the usual compact fungal mat. In Form No. 1, although radial growth was extensive, the colonies were characterized by multi-branching of the hyphae and a general sparsity of any dense mycelial formation. Hence, the comparison of the radial growth rate between fungal forms must be relative to the density of mycelial development per colony area.

Of the seven fungi studied, the greatest rate of radial mycelial growth per day occurred in Forms No. 1, 3, 5 and 6. Form No. 1 showed the most vigorous hyphal development, with some single spore isolates exhibiting as much as 4-5 millimeters of radial growth per day. Similar variation in the mycelial growth rate was observed in the different Form No. 1 isolates, both from Biscayne Bay as well as adjacent marine areas. These *Halophiobolus* fungi are being studied in pure culture on salt water agar to analyze further differences in growth rates and possible correlations with morphological variations of the different isolates. Form No. 6 was characterized by vigorous mycelium formation exceeding 2.0 millimeters per day, while Forms No. 3 and 5 produced only 0.86 and 0.71 millimeters of growth per day respectively. However, the density of the fungal mat was greater in these latter two forms than in Forms No. 1 and 6. It is significant that Forms No. 1, 3, 5 and 6, in addition to a high rate of mycelial development in artificial culture, also were characterized by a correspondingly large ascocarpic population on the salt water agar medium.

Forms No. 2, 4 and 7 exhibited a comparatively low rate of mycelial growth, together with only slight or practically negligible formation of ascocarps on salt water agar. Form No. 2 produced only 0.5 millimeters of growth per day, while Forms No. 4 and 7 exhibited less than 0.30 millimeters of growth per day. In the following discussion on ascocarpic production, the correlations between the formation of these fruiting structures on wood and in agar is presented.

Ascocarp production: Of the seven fungi isolated in pure culture, four were characterized by an extensive production of mature ascocarps, morphologically similar in the nature of the ascospore to those observed on the wood substrate (Table 3). These fungi were Forms No. 1, 3, 5 and 6, of which only Form No. 1 had a correspondingly heavy ascocarpic development on the submerged wood as well as in artificial culture. The abundant and vigorous formation of ascocarps of Forms No. 3, 5 and 6 on salt water agar was in sharp contrast with the limited occurrence of ascocarps of these forms on the wood itself.

TABLE 3
ASCOCARP PRODUCTION ON SALT WATER AGAR

Form	Time of Initial Ascocarp Development	Intensity of Ascocarp Production Per Colony
No. 1	2-3 weeks	75-100 +
2	*	—
3	2½-3 weeks	30- 50 + **
4	*	—
5	2-3 weeks	15- 20
6	3 weeks	75-100 +
7	*	—

* No ascocarps produced during period of examination, i.e., 1-12 weeks.

** As many as 100-150 in some cultures.

Form No. 3 was isolated from only two separate stations in Biscayne Bay, and at both of these areas, the visible ascocarpic production was only slight to moderate, occurring on only several panels. No large concentrations of Form No. 3 ascocarps were observed, and the ascocarpic stage of the fungus did not occupy a wide area of the exposed wood. Form No. 5 was collected from only one station on different panels several times over an 8 month period, with no more than a few scattered ascocarps on the wood surface. The ascocarps of Form No. 6, isolated at three separate stations in the vicinity of Key Biscayne, occurred in comparatively slight amounts scattered over the wood area. The distribution of ascocarps of this latter form was different from that of the other marine fungi in that the ascocarps of Form No. 6 developed superficially upon the calcium deposits of marine fouling

organisms on the wood surfaces and not on the wood itself.

In addition to variation in total number of ascocarps on wood and in salt water agar, a decided difference in time of ascocarpic maturation was noted between these two substrates, i.e., wood and agar. While the fruiting structures of Forms No. 3, 5 and 6 developed within 3 to 4 weeks on agar, similar development on submerged wood usually required from 7 to 16 weeks. Following one month growth on salt water agar, these forms developed mature ascocarps, containing viable diagnostic appendiculate ascospores. Mono-ascosporic cultures of Form No. 3 produced as many as 100-150 fruiting structures per individual colony and further transfer of the new fungal growth resulted in continued prolific ascocarp development. Similarly, Forms No. 5 and 6 showed equal rapidity in ascocarp maturation from single ascospore inocula. The mode of development or orientation of the majority of Form No. 6 ascocarps on salt water agar was different from the development of the ascocarpic stage of the other fungal forms under similar cultural conditions. The Form No. 6 ascocarps developed in nearly all cultures on the exposed glass surfaces of the culture plates, particularly along the periphery of the fungal colony on the sides of the glass culture dish. In only one culture, slight ascocarpic formation was apparent in the agar comprising the main area of fungal growth.

Since salt water agar alone is a comparatively deficient nutrient medium compared to the composite organic materials present upon, and a part of, submerged wood, it would be expected that greater and more vigorous ascocarp production would occur on the wood substrate. In Forms No. 3, 5 and 6, such was definitely not the case. Apparently, judging from the rapidity and vigor in the initiation and production of the ascocarpic stage from single ascospores in salt water agar, it is possible that greater populations of these fungi occur under natural salt water conditions than was indicated by the examination of the panels.

In Forms No. 2, 4 and 7, the opposite of the observations noted above for Forms No. 3, 5 and 6 occurred (Table 3). While Form No. 2, and to a lesser degree, Forms No. 4 and 7, developed very abundantly on the submerged wood, only a sparse ascocarpic formation was noted in salt water agar cultures of these fungal forms. Several cultures of Form No. 2 exhibited moderate ascocarp development during 8-12 weeks of incubation following mycelial inoculation. However, this

occurrence of the sexual stage was not general, and was considerably less in number and maturity than in cultures of Forms No. 1, 3, 5 and 6. Form No. 2, in particular, was characterized on the wood substrate by a dense population of ascocarps, often concentrated over the entire wood surface. The sparsity of ascocarpic formation by Form No. 2 in salt water agar becomes particularly noticeable when it is considered that the vigorous colonization of submerged wood by Form No. 2 often occurred following only 7-9 weeks of submergence. However, cultures of Forms No. 2, 4 and 7, even after several months of growth on salt water agar, did not show the ascocarpic development even remotely approaching that occurring on the natural substrate.

DISTRIBUTION OF FUNGI AND ECOLOGICAL RELATIONSHIPS

The occurrence and distribution of the various marine fungi over the area surveyed in Biscayne Bay, together with the intensity of fungal infestation at individual stations, is summarized in Table 4. While certain characteristics of the organisms themselves and conditions of the immediate ecological area may directly affect the tabular results of the survey, the areal and temporal aspects nevertheless are indicative of the occurrence and abundance of the marine fungi described. Among such influencing factors are: (1) the rate of fungal metabolism and individual growth features which have a direct bearing upon the time, rate and degree of ascocarp production. (2) the interrelationships between the fungi, whereby mycelium formation and the inception of the sexual stage in one fungus may be retarded or inhibited by antagonistic processes in another accompanying fungus. (3) the composition of the wood matrix, as well as the varied deposition of nitrogeneous and carbonaceous materials on the wood surface, and its effect upon an available nutrient supply for the fungi. (4) the chemical and physical characteristics of the area, which may directly affect spore germination or prevent the formation of the diagnostic fruiting structure, although mycelial development may be quite extensive within the tissues of the wood.

The tabulation of fungal occurrence is based upon the recognition of the ascocarpic structures on the submerged wood surfaces, and does not include potential or incipient ascocarpic production, nor the actual extent of mycelial ramification within the wood.

TABLE 4
ASCOCARPIC PRODUCTION ON SUBMERGED WOOD
FUNGAL FORMS

Stations	No. 1	No. 2	No. 3	No. 4	No. 5	No. 6	No. 7
No. 1	4	4					2
No. 2	3	4					
No. 3	5	5					
No. 4	4	5					
No. 5	5	4					
No. 6	3	4					
No. 7	3	4					
No. 8	3	4	2	3			
No. 9							
No. 10	3	3	2	2	1	1	2
No. 11	3	3					2
No. 12	3	2		2		1	
No. 13	3	2		2		1	
No. 14	3	4					
No. 15	2	4					4

SIGNIFICANCE OF NUMBERS USED IN TABLE 4

1. Scant to slight occurrence, scattered ascocarps; no concentrations, often only a few ascocarps in some cases.
2. Slight occurrence, occasional scattered concentrations of 25-50 ascocarps per square centimeter; no general distribution.
3. Slight to moderate occurrence, scattered concentrations of 50-100 ascocarps per square centimeter; general distribution.
4. Moderate to heavy occurrence, large concentrations, often 100-150 ascocarps per square centimeter; general distribution.
5. Heavy occurrence, large concentrations; general distribution (occupying entire wood surface); often as many as several hundred ascocarps per square centimeter. (Often occurring in such great density as to cover nearly completely the underlying infected wood matrix).

Observed Distribution. As reported previously (Meyers, 1953), and as indicated by the current survey, Forms No. 1 and 2 are widely distributed over Biscayne Bay, being abundant at all of the stations except No. 9. Forms No. 3-7 occur at scattered stations and in less abundance than Forms No. 1 and 2. The colonization of Form No. 7 on panels at such widely separate stations as Stations No. 1 and 15, as well as Stations No. 10 and 11, suggests that this form has a more general distribution over the Bay than indicated by the wood analysis. The absence of ascocarps of this fungus at intermediate stations may be due, in part, to the often complete biological destruction of the outer wood surfaces following long periods of submergence. Since Form No. 7 was found predominantly on older wood submerged as long as 7-12 months, such destruction would tend to remove the area of the wood containing the most abundant ascocarp population.

The examination of panels from Station No. 10 at the southwest

tip of Virginia Key revealed the colonization of all of the fungal forms collected in the survey. This station as well as Stations No. 11-13, all in close proximity to Key Biscayne, supported a more diverse fungal population, particularly of Forms No. 3-6, than at Stations No. 1-7. Similarly, the degree of wood degradation during the initial 1-2 months of submergence was considerably less at the former stations (No. 10-13) than at the latter stations (No. 1-7).

From Table 4, it is apparent that the overall concentration of Form No. 2 was noticeably greater at Stations No. 1-8 than at Stations No. 10-13. It is interesting to note that the former stations are in an area where moderate to high amounts of sewage occur, coupled with enhanced biological activity, while Stations No. 10-13 are in the vicinity of Key Biscayne, receiving considerably less direct sewage flow. Thus, enhanced fungal growth in the northern part of the Bay is similar to a recorded high rate of attachment of marine fouling organisms in this same area as observed by Smith, et al. (1950). In addition to these factors of concentration, a difference in total time required for the formation of the mature ascocarp stage of Form No. 2 was noted between those stations in the Government Cut and city dock area and those in the vicinity of Key Biscayne and the southern part of the Bay. The former area showed abundant ascocarpic development on the panels within 7-9 weeks following submergence, while the latter stations required from 13-28 weeks for mature ascocarps of Form No. 2 to appear on the wood surfaces.

Stations No. 14 and 15, in the southern Bay, showed only moderate colonization by Form No. 2 following 8-12 weeks of submergence. However, after 7 months of exposure, the panels at these two stations revealed a very heavy population of Form No. 2 ascocarps.

The study of panels and older wood submerged at Bimini, Bahamas, revealed the occurrence and vigorous population of Forms No. 1, 2 and 7 in that area. The microscopic examination of these fungi indicated that Forms No. 2 and 7 apparently are similar morphologically to those species isolated from wood in Biscayne Bay. The appearance of these three marine fungal types in Bimini that have been collected in Biscayne Bay, suggests the possibility that the additional fungal forms observed in Biscayne Bay may be collected at a later date on wood at Bimini.

The extensive colonization of Form No. 2 at Key West and Bimini, Bahamas, establishes this fungus as a halophilic organism of wide geographic distribution. In addition, a recent examination by the writer

of wood submerged in Panama Bay, Canal Zone, by Dr. Donald D. Ritchie of the Tropical Exposure Laboratory, ascertained the occurrence of Form No. 2 in that area. The species of Form No. 2 from Panama appears similar in ascocarpic character and ascospore size to the fungus collected in the Florida area.

Further evidence of the occurrence in other marine areas of fungal forms currently found in Biscayne Bay is indicated by the examination of wood from Port Aransas, Texas, Cortez, Florida and Cedar Keys, Florida. These latter two areas on the Gulf Coast of Florida revealed the colonization of Form No. 2 on submerged wood, while wood from the Institute of Marine Science, Port Aransas, Texas, showed ascocarps of Form No. 3. The isolation of Form No. 3 is the first recording, other than the New England area, of the occurrence of this fungal form outside the Biscayne Bay area.

The study of panels from areas other than Biscayne Bay, e.g., Panama, Key West, Texas, Mississippi, Cedar Keys, Cortez, Dunedin (Florida), all have revealed the occurrence of different types of Form No. 1 on submerged wood. Variation in ascocarpic size and length of the appendages among the different *Halophiobolus* isolates indicates that the fungus is of wide ecological range in subtropical and tropical marine waters. Analysis of the characteristics of the different Form No. 1 types is in progress to ascertain the validity of certain morphological features for future separation of species.

Ecological Relationships. The examination of the wood panels showing various degrees of degradation revealed abundant marine fungal activity accompanied by other aspects of the ecological complex. The observations reported below represent a compilation of analyses of wood panels exposed at the different stations throughout the investigation period. Although the Form No. 2 organism is used as the representative fungus, in view of its wide and abundant occurrence and vigorous growth rate on wood, similar correlations may apply equally as well to the other fungal forms.

(A) A softening or dissolution of the outer cell layers of the submerged wood surfaces was accompanied, in nearly all instances, by various intensities of a marine fungal population. The first fungi to appear on the submerged wood were usually the various *Halophiobolus* forms (Form No. 1), closely followed in occurrence by Form No. 2 ascocarps. As reported in the earlier publication, these two fungal forms were often associated together on the submerged wood surfaces.

The appearance of a vigorous ascocarpic population early in the

submergence of the wood precludes the earlier development of the mycelial stage of the fungus throughout the infected wood tissue. Thus, it is evident that fungal degradation processes are occurring in the submerged wood well ahead of the recognition of the fungus on the wood substrate as mature ascocarps. This ramification of the fungal hyphae, comprising the mycelial stage, enables the organisms to invade the underlying wood tissue, often with accompanying dissolution of the wood components.

The initial "conditioning" of the outer wood matrix exposed to the continuous or intermittent action of sea water is apparently preliminary to more extensive damage by other biological or mechanical agencies. In connection with *Teredo* attack on newly submerged panels, Isham and Tierney (1953) postulated that "either the effects of the surface microflora or of the sea water itself, or a combination of both, is responsible for the softening and partial removal of the intercellular material of the wood." The present study of wood panels submerged in Biscayne Bay definitely establishes that marine fungi are an abundant and continuous biological component of the surface microflora on such wood. In addition, the vigorous penetration of submerged wood by marine fungi and the cellulose and lignin-decomposing activity by these halophilic organisms (Barghoorn, 1944) indicates that wood and wood materials afford a normal substrate for this group of marine fungi. While marine fungi may not be the only organisms instrumental in the important *early dissolution* of the outer surfaces of submerged wood, it appears that they have a significant part in such biological degradation processes.

(B) While attack by *Limnoria* and *Teredo* constitute the more obvious damage of wood submerged in salt water, extensive colonization of such wood by marine fungi may occur occasionally *without* extensive marine borer infestation. This was noticeable especially in the case of Form No. 1 where ascocarpic formation occurred as early as 4½-7 weeks following submergence. In addition, during the early phase of the exposure period, i.e., 6-9 weeks, the panels at Station No. 3 supported a dense ascocarpic growth of Form No. 2 with comparatively slight wood borer infestation. This intense fungal colonization comprised as many as 150-200 mature ascocarps per square centimeter over the entire area of the inner wood surfaces.

The majority of exposed panels examined showed the association of marine fungi, particularly Forms No. 2 and 7, with marine borer damage throughout the submergence period of the wood. However,

one series of panels submerged for 7 months at Station No. 15 revealed a large population of Forms No. 2 and 7 with only moderate accompanying marine borer activity. The panels at the different stations undergoing extensive attack by marine borers showed abundant formation of the Form No. 2 ascocarps within the borer cavities and in the inner matrix of the eroded infected wood tissue. Thus, it appears that vigorous fungal growth may occur on submerged wood under conditions of *negligible* marine borer activity as well as *before* extensive damage of the wood by these marine animals.

It is significant that in other marine areas, i.e., Bimini, Bahamas and Key West, similar observations were recorded. A very large degree of marine borer activity at Bimini was paralleled by a very extensive development of Forms No. 2 and 7 well within the eroded wood. On panels submerged at Key West, a large population of Form No. 2 on the wood following 7-9 weeks of exposure preceded any extensive degree of wood borer activity.

(C) Although all of the marine forms isolated appear to be normal wood-inhabiting fungi, the production of ascocarps in artificial culture, devoid of any wood source, suggests that the absence of wood in the natural environment is not a limiting factor for occurrence and development of these fungi. The area where the different stations were located is, in general, well supplied with sources of older wood, i.e., pilings, piers, docks, and other wood remains, so that an exact analysis of the influence of adjacent wood on fungal occurrence and abundance cannot be made. The highest concentration of fungi was at Station No. 3, a metal bouy approximately 150 yards from any submerged wood. However, the appendiculate nature of the ascospores of these fungi indicates that they are capable of ready distribution within an aquatic area, and are not necessarily confined to a particular sector of that area. Hence, the effect of an abundant source of fungal-infected wood in one area may reveal itself in a vigorous fungal population in adjacent and scattered areas, all within the range of similar water movement.

While water movement facilitates the dispersion of the spores of the fungi, an increased current flow does not appear to decrease the attachment of the fungi to the submerged wood surfaces. The intensity of fungal infestation at Station No. 3, subject to continuous rapid tidal flow through Government Cut, showed no decrease in the attachment of marine fungi regardless of the constant water flow. In fact, the analysis of the submerged panels revealed a greater degree of fungal

colonization than at nearby Stations No. 1 and 2, where the submerged panels were not in a direct current flow as at Station No. 3. The effect noted by Doochin and Smith (1951), whereby an increased water flow reduces the intensity of marine borer attack, apparently does not apply in the case of these marine fungi.

(D) The analysis of the exposed wood panels revealed a definite variation in the abundance of visible ascocarps on the southern yellow pine panels as contrasted with the northern spruce wood. The yellow pine wood supported a greater population of ascocarps of the different fungi described, as well as exhibiting less rapid surface erosion than the northern spruce wood. The contrast in density of ascocarpic formation between the two woods may be due in part to the different anatomical structure of the woods. Northern spruce is, in general, a lighter wood of more uniform texture with less summer growth than southern yellow pine, which has a larger development of summer wood. It is this latter wood tissue that retains its rigidity during the early stages of submergence, while the spring wood undergoes more rapid softening and disintegration. Hence, while fungal activity may be more vigorous in early stages of the destruction of the northern spruce surface, the resultant accumulation of ascocarps may be prevented by the continuous sloughing off of the young ascocarps concurrent with that of the outer infected wood.

It is expected that current, as well as contemplated, physiological experiments involving these halophilic fungi will clarify the various distributional data collected in the survey. During the course of present studies, it is apparent that laboratory and field measurements of fungal growth rates offer a more complete picture of the fungal phenomena than does laboratory or field analysis alone. The correlation of the various aspects of these two habitats, i.e., salt water agar and the natural salt water environment, with marine fungal activity, will be continued in conjunction with the basic nutritional studies of the organisms themselves.

REFERENCES

- BARGHOORN, E. S. and D. H. LINDER
1944. Marine fungi: their taxonomy and biology. *Farlowia*, 1 (3): 395-467.
DOOCHIN, H. and F. G. W. SMITH
1951. Marine boring and fouling in relation to velocity of water currents. *Bull. Mar. Sci. Gulf and Caribbean*, 1 (3): 196-208.
ISHAM, L. B. and J. Q. TIERNEY
1953. Some aspects of the larval development and metamorphosis of *Teredo* (*Lyrodus*) *Pedicellata* De Quatrefages. *Bull. Mar. Sci. Gulf and Caribbean*, 2 (4): 574-589.

MEYERS, SAMUEL P.

1953. Marine fungi in Biscayne Bay, Florida. Bull. Mar. Sci. Gulf and Caribbean, 2 (4): 590-601.

SMITH, F. G. W., R. H. WILLIAMS, and C. C. DAVIS

1950. An ecological survey of the subtropical inshore waters adjacent to Miami. Ecology, 31 (1): 119-146.

ZOBELL, C. E.

1946. Marine Microbiology. Chronica Botanica Company.

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TABLE OF CONTENTS

	PAGE
1. ILMO HELA: The surface current field in the western part of the North Atlantic	241
2. RUTH E. LENDERKING: Some recent observations on the biology of <i>Littorina angulifera</i> Lam. of Biscayne and Virginia Keys, Florida	273
3. JOHN C. AYERS: A method for rendering wood resistant to marine borers	297
4. NORMAN MILLOTT: A note on the shedding of pigment from the body wall of <i>Holothuria grisea</i> Selenka	305
5. SAMUEL P. MEYERS: Marine fungi in Biscayne Bay, Florida. II. Further studies of occurrence and distribution	307